

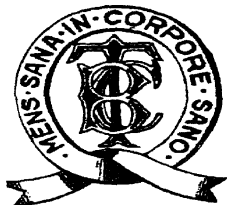


# THE PENICILLIA

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*preparing the illustrations*



LONDON  
BAILLIÈRE, TINDALL AND COX  
8 Henrietta Street, Covent Garden, W.C. 2  
1930



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**PRINTED IN AMERICA**

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## INTRODUCTION

The molds of the genus *Penicillium* share with the *Aspergilli* and the *Mucors* a noisome preëminence as weeds. They rot our fruit, attack our vegetables and meats, injure our stored grain, spoil our soft drinks and our bottled water, contaminate our pantries and kitchens, and even attack our bodies. They infect and at times destroy the usefulness of solutions and moist precipitates, discolor fibers, wood, paper stock, stored paper and sometimes our books. In the laboratory they infest and often invalidate every kind of culture operation, bacteriological, mycological, or phanerogamic. To offset these activities the chemists have gathered a little return by using them in biochemical investigations and the cheese industry has capitalized their enzymic activity to ripen such cheeses as Camembert and Roquefort. Otherwise their possibilities of usefulness remain mostly unknown, but their presence is thrust upon us so frequently that some means of identifying them is very desirable.

This study of *Penicillium* goes back to 1904 when the author, assigned to cheese investigations at the Storrs Agricultural Experiment Station by the United States Department of Agriculture, was first compelled to attempt to identify the molds active in the cheese cellar. With that end in view, he carried a miscellaneous lot of *Penicillia*, *Mucors*, *Aspergilli*, *Fusaria*, *Trichodermae* and the nondescript forms commonly called *Monilia*, *Oidium*, *Oospora*, etc., to the Cryptogamic Laboratory of Harvard University for study. Prof. Roland Thaxter, passing his desk, looked the lot over, came upon the various unrecognizables, and said approximately, "Why bother with these miserable little things? Why not do something worth while, straighten out *Penicillium*? It is an awful mess." The challenge was accepted and has been kept in sight through many vicissitudes which have at times taken him far afield during these twenty-five years. At best, it is possible to present only a very unsatisfactory scheme of classification of this difficult group.

In his first scheme of study (Thom 1905) he attempted to follow the lines set by bacteriology in proposing to separate species upon the presence, absence, or intensity of selected biochemical reactions. This proved very useful in helping fix upon fairly broad groups, but, within methods at that time deemed practical, it failed to furnish a satisfactory

separation within related series. Carried to a quantitative chemical basis, some such scheme remains the only hope of fixing the individual strain, but this service can only begin when the resources of the morphologist in the culture room have determined the group involved.

Even in the publication of his "Cultural Studies" (1910), he retained the futile hope of a cultural and morphological species-diagnosis which would make possible the identification of every *Penicillium*. By 1914, this was abandoned for the delimitation of groups presenting common morphological and cultural characters. In these groups, forms obviously morphologically related are brought together and the contrasting characters are mainly quantitative: the same structures a little larger or a little smaller. The same reactions, partially suppressed or variously accentuated, often give marked contrasts making for individuality in the strain or species, but frequently, in a series of cultures, merge so completely that the task of diagnostic description becomes endless. This was further developed in the study of the *Aspergilli* (1926) and is followed here.

Some 600 names have been found applied to *Penicillia* and related organisms. Although a considerable number of these "species" are bibliographic changes in nomenclature without evidence of actual study of the molds themselves, more than 400 of these names were proposed by authors with actual material before them. We know the organism described in somewhat less than half of these 400 species, and even among these the purity of the type used is often questionable. The bodies of these fungi are delicate and easily crushed beyond recognition; when dry they fall to powder under very slight pressures and a breath blows the powder away.

Alive and actively growing, they have individuality as pronounced as their capabilities for evil, but the elements of that individuality, color, odor, and habit of growth, are as evanescent as frost designs on a window pane in winter. To lay a foundation for a permanent knowledge of this lot of molds, the whole range of morphology and physiology must be searched for marks of separation stable enough, and sharply enough marked to convey to the reader a definite picture of the organisms studied. Then organism by organism they must be fitted into the scheme of classification to form a consistent and interpretable whole. That ideal has not been reached, but we hope that a tangible outline has been made and enough types established to insure group recognition at least for the more abundant molds of the penicillate series.

With 600 names to pick from, the description of organisms as new

species might not seem to be necessary. No new species appear in our notes from the date of the publication of Biourge's monograph until the chapters of this book were actually organized for publication. It was then found necessary to account for particular combinations of colony structure and type of penicillus in a number of places, and to propose new combinations where names proposed by others had been previously used for species published as *Penicillium*. It is entirely probable that other combinations of characters will be found which will add materially to the list as given. It is hoped that a framework has been presented into which such forms may be easily fitted.

Such a grouping system as this necessarily involved constant readjustment on the part of the writer. Notes and descriptions five years old were thus frequently inadequate when the groups were finally put together. Hundreds of records of organisms studied, were discarded, and many forms in the collection for periods of ten years, were ignored for sheer lack of time to work them over again in detail. Occasional identifications for correspondents, or cultures distributed under names, will probably fail to fit the scheme of classification presented. This is unfortunate but incident to a developing study of such a nature.

The task of identifying a species of *Penicillium* will not be easy even with the descriptions and keys presented in this book. If an organism is sufficiently important, or sufficiently interesting, to be worth studying, the worker must know it well enough to determine where it belongs in some orderly scheme of classification before he can correlate his own findings with the work of others. Making such a study of an organism insures a picture of the species in the mind of the investigator, definite enough to protect his subsequent culture studies against loss, contamination of his species, or the substitution of another form. If the complexity of the task of identification proposed here, deters some<sup>1</sup> from the use of Latin generic and species names for undetermined green molds, the author gladly accepts the responsibility.

The author is keenly aware of the deficiencies in this book. He has barely mentioned the possibilities of the genetic line of attack indicated by the work of Derx and more recent work of Biourge as the true basis of a permanent nomenclature of these forms among the ascomycetes.

<sup>1</sup> In writing this we have in mind an elaborate piece of biochemical work upon a particular activity of "*P. glaucum* Link." The chemist when asked for his culture confessed he had lost it and referred us to the one who furnished it to him who replied, "——— asked for a green *Penicillium*, we will guarantee that the culture was a *Penicillium* and that it was green—that's all we know about it."

Nevertheless, he is confronted with the need of a cultural and morphological scheme for practical identification of strains as they are encountered in the laboratory and in industry. *Penicillia* met in such situations are nearly all hyphomycetes (reproduce only by asexual spores). Within the time and experience of the general worker, ascospores are rarely obtained and then only in a few selected species. A scheme of nomenclature which will enable a worker to locate his form within a fairly homogeneous group in which one or more species have been described, will open up to the investigator the existing literature dealing with his organism or its near relatives. Such organisms should be placed as closely as possible in a scheme of classification; their characteristics and morphology defined as fully as possible, and in cases of great industrial or biochemical importance should be preserved as multiple cultures in the hands of specialists in several different laboratories to insure their availability to other workers.

The obligations incurred during these twenty years include members of many laboratory groups and many individual students in America and in distant lands. Only a few, including those who have contributed largely to our collection and our information, may be mentioned here. Prof. Ph. Biourge of Louvain contributed his entire collection after the publication of his monograph in 1923. Dr. Johanna Westerdijk and her assistants at Baarn furnished us the Zaleski collection and many individual cultures. In addition, we must mention the late Prof. G. F. Atkinson, followed by Professor Whetzel and their associates at Cornell; Professors Farlow and Thaxter at Harvard; the mycologists of the New York Botanical Garden and the Director, Dr. N. L. Britton, who put its resources at our service; Prof. Carl Wehmer at Hanover; Dr. R. Westling at Stockholm; Miss E. Dale at Cambridge, England; Mr. W. B. Brierley at Rothamsted; Mrs. M. N. Kidd of London; Mr. H. Raistrick and his associates in Ayreshire, Scotland; Mr. F. M. Putterill at Cape Town; Mr. P. A. Van der Bijl at Pretoria; Mr. C. G. Hansford in Jamaica; Dr. O. daFonseca in Rio de Janeiro; Drs. Saito and Naganishi at Dairen, Manchuria; Prof. Jun Hanzawa at Sapporo; The British Cotton Industry Research Association; and many colleagues in the various bureaus of the United States Department of Agriculture who have contributed freely of their isolations in this group.

The completion of the work has been greatly aided by the collaboration of Dr. O. E. May of the Bureau upon the biochemical phases of the work, and Dr. M. A. Raines in preparing many of the illustrations.

Finally, Dr. Margaret B. Church carried the burden of bibliographical

work, of correspondence, of the culture collection and of the culture room for ten years with discriminating judgment. Without that help the task would hardly have been done, because many thousands of cultures were made, recorded and examined each year. Nevertheless in the classification of *Penicillia*, the mistakes and errors are the author's, not hers, and the author is solely responsible for the book aside from certain sections in which Doctors Church, May and Raines are credited in the text.

CHARLES THOM.

*Washington, D. C., 1929*





## CHAPTER I

### CHARACTERIZATION OF THE GENUS *PENICILLIUM*

By common consent the generic name *Penicillium* proposed by Link in 1809 has been accepted for a group of hyphomycetes producing conidial fructifications in the form of a brush or broom called from the Latin term, the penicillus (fig. 1). The name has been loosely applied to all molds having conidial apparatus suggestive of such a brush or broom. Many times in the descriptions published this application of the name has rested upon superficial resemblances rather than actual homologies, hence members of several different genera have from time to time been given specific names as *Penicillia*. Close scrutiny of the historical use

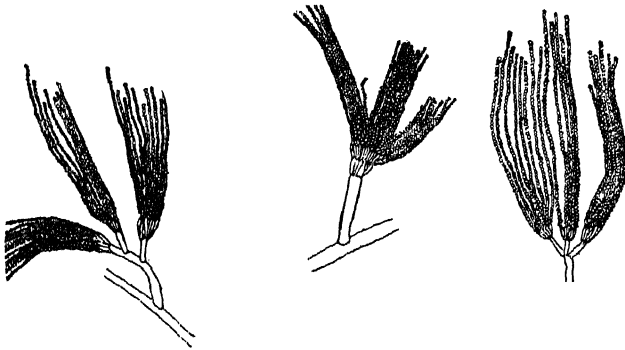


FIG. 1. The penicillus: three "brushes."

of the name as outlined in another chapter (Chapter II) convinces us that one of the three species originally described belonged to the series producing the universal and destructive *Penicillium* rot of apples in storage. We may, therefore, accept the type of morphology encountered in this species, *P. expansum* Link as emended by Thom in 1910, as a basis for characterizing the genus. Since the name is based upon the conidial apparatus presented in the form of a brush or broom, the penicillus, the production of this apparatus must be clearly understood.

#### LINK'S TYPE

Since *P. expansum* Link (compare no. 315) is proposed as the type of the genus, it is perhaps well to introduce the original description from

Link's "Observationes," 1809, p. 17, under division *c* of generic discussion.

*c.* Floccis aliis ramosis complicatis, aliis simplicibus erectis.

*P. expansum.*, floccis albis, sterilibus magis minusque contextis, capitulis sporidiisque glaucis. In fructibus putridis velutum tenue format, in fungis aliisque corporibus putridis crassius, in corporibus saccharo conditis membranas e floccis valde contextis. E membrana haece, quae revera Mucedinis pars est emergunt flocci teneri, albi, capitulis glaucis. An *Monilia digitata* Pers?

Since much confusion was introduced by the change to *P. glaucum* introduced by Link in 1824, the description of *P. glaucum* in the "Observationes" is also given.

*a.* Floccis simplicibus.

*P. glaucum*, caespitibus effusis, floccis albis, capitulis sporidiisque demum glaucis. In corporibus putridis frequens. Maxime affine *P. expanso*, et forte ipsius varietas nondum perfecta. Iconem v. fig. 24.

#### THE CONIDIOPHORE

The conidiophore or fertile hypha is a branch arising from a vegetative hypha which is either submerged in the substratum or forms part of some sort of mat or felt upon or above the surface of the substratum. This fertile hypha usually shows little or no differentiation from the vegetative hypha from which it arises as a branch. It stands commonly, but not always, perpendicular to the long axis of the main hypha. In a few series, the conidiophore is larger in diameter than the vegetative hypha. In some series, the conidiophore walls show more or less conspicuous pitting, so abundant and prominent as to give the surface a distinctly rough appearance, in others such pitting while present can only be seen by careful use of high magnifications; in others, no pitting is determinable. Warts, granulations or encrustments are present in particular species. In some cases, these encrustments are soluble in water, alcohol or both, hence are lost in the preparations commonly studied.

For descriptive purposes, the length of the conidiophore must be regarded as the distance from its point of origin in a vegetative hypha to the base of the branching system or penicillus. It may be simple or unbranched, or variously branched. It may stand alone, or be more or less closely aggregated into clusters, tufts, fascicles, ropes or definite coremia with stalk and head, or may arise as a branch from a similar aggregation of apparently vegetative hyphae.

THE *PENICILLUS*

The penicillus (English, brush or broom; German, pinsel) is understood to cover the whole branching system (fig. 2). The measurements, however, as commonly reported for this system are exclusive of the chains of conidia since there exists some degree of uniformity, or regularity of diversity within the species as to branching, whereas the chains

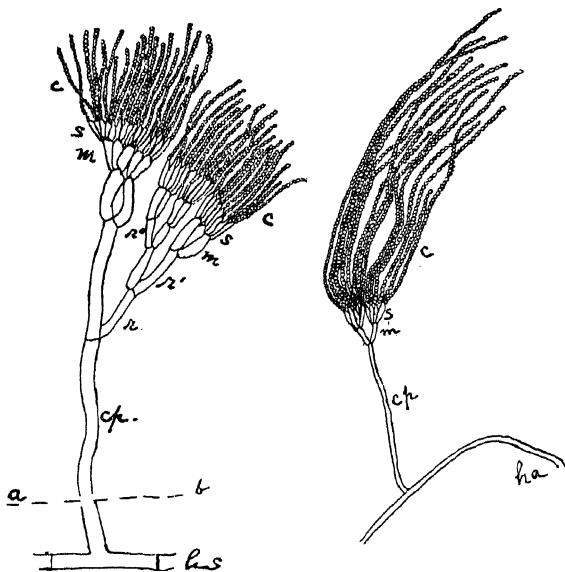


FIG. 2. The penicillus: *ab*, the surface of the substratum; *c*, the conidia; *cp*, conidiophore; *ha*, aerial hypha; *hs*, submerged hypha; *m*, metulae; *r*, branches or rami; *r'*, branchlets or ramuli; *r''*, secondary branchlets; *s*, sterigmata.

of conidia may reach great lengths in the old culture without affecting the measurements of the fruiting structure. For purposes of discussion, our consideration of the elements in this branching system will begin with the terminal cells (sterigmata) which bear the conidia, since these are common throughout the group, and will progress backward toward the main conidiophore.

## THE STERIGMA

The differentiated conidium producing cell, characteristic of *Penicillium* and related genera is variously named the sterigma (fig. 3), (plural,

sterigmata), conidiiferous cell or basidium. Biourge uses the term phialides for the sterigmata. Since the term sterigma has been rather widely accepted, it will be used here in spite of what seem to be fairly strong theoretical objections. The sterigma is a transformed cell with a tubular body of fairly typical length and diameter characteristically narrowed at the tip to a conidium producing tube from which conidia are cut off successively by cross walls.

The first sterigma of a verticil or group is a terminal cell which becomes transformed into a spore producing organ, the second buds out at the base of the first and successive sterigmata bud out to form a whorl, verticil or cluster, on the apex of the main axis or stalk. The apex of the stalk may be unchanged in size or variously enlarged toward the appear-

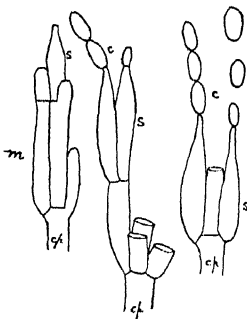


FIG. 3. The sterigma (*P. pinophilum*): *c*, conidia; *cp*, conidiophore; *m*, metulae; *s*, sterigmata.

ance and proportions of the vesicle of an *Aspergillus*. The number of sterigmata in the verticil may be few and readily countable, but in most species becomes large with such crowding as makes the fixing of characteristic numbers entirely impracticable. Aside from an exceedingly small number of species, numbers of sterigmata as given in species descriptions are misleading.

The French group including Gueguen, Vuillemin and Bainier have regarded the successive production of the sterigmata in the verticil as distinguishing *Penicillia* from *Aspergilli* in which the members of the verticil of sterigmata are produced simultaneously. Bainier and Sartory describe (Bul. Soc. Myc. France, 28 (1912), p. 48, plate I) and figure *Citromyces* as producing first a terminal sterigma followed by the remainder of the verticil in series from the apical cell which may enlarge to

a vesicle-like organ as they develop. *Aspergillus* is figured as producing a vesicle with a whole group of sterigmata simultaneously developed.

#### CONIDIUM FORMATION

The method of conidium formation was discussed by Thom in 1914 and more recently (1928) by Scaramella. Thom showed that the newly formed conidium is at first a cylindrical segment cut from the conidium bearing tube at the tip of the sterigma. It attains the size and shape characteristic of its species by lengthening and swelling, and by the formation of its own cell wall within a primary wall which may be regarded as derived from the parent cell or sterigma. The new wall or true spore wall may blend with the primary wall to leave no visible



FIG. 4. *Penicilli*: A series increasing in complexity from monoverticillate to triverticillate.

line of separation, or may only partially combine with the old wall, leaving a bridge or connective at times easily visible between the conidia in the chain. Again, also as in *Aspergillus*, there may be a wrinkling of the old wall to form ridges or spinulose points on the conidium with or without some deposit of coloring matter or other substance between them to give coarseness or body to the roughenings or spinulosities.

In Brefeld's original work the formation of the conidium was regarded as a budding process comparable to the multiplication of yeasts except for the specialized point at which the budding occurred. Another view, based upon the original discussion by Guéguen (1899), regarded conidium formation as "endogenous," the separation of a spore mass within a mother cell in such a way that it was allowed to slip out through a tube. The theoretic difference as seen in Scaramella's figures is practically confined to the cell wall relations without seriously affecting the observations of either group which agree in the general morphology represented in the cell divisions reported.

Conidia once formed may remain connected in chains of a few to 200 or more, or may fall away quickly. These chains of conidia may diverge widely, become a tangled mass, stand almost rigidly parallel or be closely aggregated into columnar masses. These masses may even become aggregated into crusts covering large areas of the colony. In any case, the same general type of arrangement of the conidial mass is recognizable in successive cultures for each species studied.

In its simplest form then, the penicillus (fig. 4) is a single verticil or cluster of sterigmata on the tip of a fertile hypha and each sterigma produces an unbranched chain of conidia. This simple form is actually found in a few species of the monoverticillate group, showing little if any irregularity. Other and manifestly nearly related forms show simple monoverticillate heads and other heads or penicilli with an occasional secondary branch or more at the next node or septum in the conidiophore, bearing a penicillus closely resembling the primary penicillus.

#### METULAE

These secondary branches appear upon a very small percentage of the conidiophores in some species, become common in others without losing the character of merely accessory fruiting branches, then merge through a series of forms showing all gradations, into characteristic branching systems. At first the branches in the cluster of branches are unequal in length and diverge without particular arrangement; then forms are found in which the sterigmata are supported by a second verticil of well differentiated cells (rarely septate) which have been called branches, basidia, secondary sterigmata and finally metulae by Westling in 1913. The term metula, previously unused for similar structures, has been accepted by recent authors and will be used here.

If we accept the sterigma as the primary conidium-producing organ, the term metula may properly be restricted to apply to characteristically differentiated and symmetrically developed members of the second series of branches, occurring in verticils and each supporting a verticil of sterigmata. This would exclude the use of the term for undifferentiated branchlets producing verticils of sterigmata (Chapter XIII).

#### BRANCHES (RAMI IN BIOURGE)

In the more complex types of penicillus such as that of *P. expansum*, the main conidiophore bears one or more series of branches which may be alternate, opposite or incompletely verticillate, but are more or less characteristically arranged in each of the groups of species represented.

In addition to monoverticillate and biverticillate types, there arise in this way polyverticillate series of species with distinguishing characters based partly, at least, upon the morphology of the conidiophore and penicillus.

In the polyverticillate group as worked out by Vuillemin, Bainier and others, symmetrical verticils of elements appear at every stage of branching so that the penicillus may show several superposed series of such branches.

#### MYCELIUM

The vegetative mycelium in *Penicillium* is a complex network of septate branching thin-walled hyphae. In many species the entire vegetative mass is submerged in the substratum. The surface growth in these forms consists of the fruiting stalks, or conidiophores only, which rise like the stems of a field of wheat (described as velvety or velutinous). Such submerged mycelium is usually massed within the first few millimeters below the surface. Some species, however, have been found to penetrate deeply into the mass infected. Another group of species produces a mass of branching hyphae variously disposed above the surface of the substratum. These aerial hyphae may creep on the surface or loop partly below, partly above the surface. Again they may be piled into cottony, floccose masses or form long tangled (lanose) woolly areas, or be variously combined into procumbent or ascending fascicles or ropes from which the conidiophores arise as branches. Vegetative hyphae in *Penicillium* are usually colorless or bright colored and often produce bright colors in the substratum itself. Brown or black masses are found in some species, but the cell walls remain uncolored. Brown walled types are not regarded as *Penicillia* in the strict sense, although some brown walled forms with penicillate fruiting masses have been described and one or two of them are discussed here.

#### SCLEROTIA

Definite pseudoparenchymatous masses or sclerotia are found in certain groups of *Penicillia*. In our experience these forms have entirely failed to become ascogenous, although Brefeld and Schwartz have reported the slow formation of asci in these masses. For descriptive purposes their presence and distribution are significant accessory information.

#### PERITHECIA

Ascosporic *Penicillia* have been described by a number of authors (Brefeld, Morini, Zukal, Wehmer, Klöcker, Dangeard, Thom, Lehman,



Bezssonoff, Derox, Biourge, Schwartz). Perithecia are regularly produced by certain species in the biverticillate series. Recently Derox has reported that perithecia in that group are produced by the union of unisexual strains. This would account for the extreme rarity of such structures even in the group in which they occur most frequently.

Theoretically a sound classification of the Penicillia can only be based upon ascospore formation. Practically the results already attained among the Penicillia confirm the experience of Blakeslee with the Mucors and Dodge's work with "*Monilia sitophila*" in showing that the strains combining in the production of ascospores are so closely related in conidial morphology that a thorough study of the grouping of these conidial strains is practically essential as background for the experimentation necessary to produce the perithecial stages.

The production of an ascosporic form is so conspicuous a character when found, that the observation leads quickly to identification. But the student of mold distribution and activities must find a basis for a temporary but fairly stable nomenclature in the non-ascosporic structures regularly found, as a basis for discussing the practical or technological problems involved. He can not wait for ascospore formation before describing his mold or seeking in the literature for previous work with the same species (compare Chapter VI).

As a convenient summary of this discussion, the following characterization of the genus *Penicillium* is offered:

#### GENERIC DIAGNOSIS

*Penicillium*. Vegetative mycelium abundant, entirely submerged or more or less effused, monopodially branching, septate, commonly showing vegetative anastomoses, colorless or secondarily colored by products of metabolism which frequently also discolor the substratum, never with hyphal walls brown or dematiaceous; colonies green, yellow green, blue green, gray green, or less commonly colorless or in avelaneous to yellow, reddish, purplish or other shades, frequently discolored in age even with spores brown in mass, but not dematiaceous; fertile hyphae or conidiophores arising as branches from the vegetative mycelium, frequently perpendicular to the vegetative hyphae, but not showing differentiated foot cells as in *Aspergillus*, with walls in some forms smooth or undifferentiated, in others more or less conspicuously pitted or roughened from secondary thickening, not dematiaceous; conidial apparatus forming a brush or broom, the penicillus, ranging from a single terminal verticil of conidia-bearing cells or sterigmata, or, a terminal

verticil of equal branches or metulae bearing verticils of sterigmata, to complex branching systems ending in verticils of specialized cells or metulae bearing verticils of sterigmata; sterigmata bearing single unbranched chains of conidia each cut off as a cylindrical segment from apical, straight, conidia-bearing tube; conidia cylindrical to oval, elliptical or commonly finally globose, smooth or roughened, colorless or variously colored, especially in mass, but not dematiaceous when examined with the microscope.

Sclerotium or perithecium formation known only in certain species without thus far definite establishment of generic types based upon these structures.

## CHAPTER II

### HISTORICAL

Species of *Penicillium* are so abundant and so conspicuous in all sorts of stale or decaying organic matter that they constitute a part of the common conception of mold and are generally called "blue" mold or "green" mold. It is easy to believe, therefore, with Brefeld that some *Penicillium* furnished the material for *Aspergillus albus* in figure 3, table 91, of Micheli's "Nova Plantarum Genera" in 1729 (fig. 5). Again, it is common mycological tradition that *Mucor crustaceus* of Linnaeus (1753, 1763) was some *Penicillium*. This name passed from author to author without added information until Persoon, who appears to have included these species in his conglomerate genus *Monilia*. It probably reappears in *P. crustaceum* Fries but there is no continuity in materials. Saccardo went so far as to cite *Monilia digitata* Persoon as the basis of *P. digitatum*, in the *Sylloge* in 1880, although he furnished little proof of the continuity of this view even as tradition. Bulliard used the name *Mucor penicillatus* for these "broom" or brush-like forms. Link, in 1809, taking the name from the specific name used by Bulliard (fig. 6), proposed the generic name *Penicillium* and applied it to three species, basing his description rather upon the morphological effect of a brush or broom than upon a definite spore producing mechanism as is shown by his handling of the genus in 1824. Link (1809) applied this generic name to three species, *P. glaucum*, *P. candidum* and *P. expansum*. Close scrutiny of these descriptions (see Thom 1910) discloses no clue to the forms studied as *P. glaucum* and *P. candidum*. *P. expansum*, was, however, indicated as common upon rotting fruits. In a survey of the native fruits on the market in Berlin (made by Thom in 1905) the same species was found to be the common *Penicillium* rot of apples, *Mespilus*, etc., in that city, as is usual in America. Review of Wehmer's (Beitr.) study of fruit rots points to the same species, apparently regarded by him at that time as *P. glaucum*. Thom accepted this species as *P. expansum* Link and regarded it as the type of Link's genus, since it is the sole species described by Link, which is recognizable with a reasonable degree of certainty. Link, in 1824, abandoned *P. expansum* and called all the green *Penicillia*, *P. glaucum*. This practice has been followed by many workers to the present day with the result that the use of the name

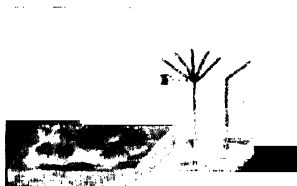


FIG. 5

FIG. 5. Micheli's Taf. 91 figure 3, *Aspergillus albus*, is reproduced to show our slender basis for identification of older species.



FIG. 6

FIG. 6. Bulliard's plate 504, figure 11, *Mucor penicillatus*, gives a better basis for identifying.



*P. glaucum* merely indicates that the organism referred to is a green *Penicillium*, but gives no further clue to identity. Another group find their type for the genus in *P. crustaceum* Fries which most of them assume to be identical with *P. glaucum*. Our recent studies of large numbers of cultures point to a different conclusion. *P. expansum* and its close allies do not produce conidia which break off from the surface of the colony in crusts, whereas several members of another series, present exactly the picture suggested by Fries. Since it is not possible to determine in any satisfactory manner which species is Fries' organism *P. crustaceum*, the name may best be abandoned. Again Biourge finds the type of the genus in *P. leucopus*, which he traces to Persoon. Biourge's organism which we have in culture (4733.81) is one of the strains difficultly if at all distinguishable from Thom's *P. expansum* which Biourge, judging by his discussion and by the culture returned to us (no. 4733.55), failed to interpret correctly. We see no reason to abandon the name selected by Thom in 1910 for this form, hence it appears here as *P. expansum* and as representing Link's idea of the genus as closely as can be determined.

The same group of species with minor variations in description passes from author to author until Corda's beautiful detailed figures of this genus appear between 1837 and 1839. In *Penicillium* as in *Aspergillus*, Corda cleared up many of the uncertainties of observation so evident in previous discussions of the genus, but idealized his details of species morphology to such an extent that identification of even one of them has never been satisfactory. Species of *Penicillium* are found described in the works of Montagne, Bonorden, Fresenius and Preuss during the period about 1850. These authors included with what we now call *Penicillia* such organisms as *Cladosporium*, *Hormodendrum* or even *Monilia sitophila*. Few if any of the names, they proposed, can now be recognized as *Penicillia*. These workers represent a period in which the mycologist was primarily a microscopist intent upon describing the specimens which came to hand either as fresh materials from his own environment or as herbarium specimens. A necessary corollary to his work was the assumption that a mold found in any situation was sufficiently pure and characteristic to form a safe basis for taxonomic work. No cultures were made. The reactions of one organism to the presence of one or several others was not taken into account. The specimen was described, then labeled and dried for the herbarium. The description was published whether the specimen was recognizable after it was preserved for the herbarium or not.

The same difficulty is encountered in interpreting the species of *Peni-*

cillium described by Rivolta, Berkeley, Berkeley and Broome, Spegazzini, Cooke, Ellis, Morgan and even Saccardo, all of whom were active, primarily as collectors, and brought to light many species of real value among groups which make more satisfactory herbarium specimens.

Constructive cultural study of *Penicillia* begins with Brefeld, whose study of the life history of *Penicillium glaucum* was published in 1874, although some recognizable work with cultures preceded that date. He limited the discussion to one species, *P. glaucum*, which is nowhere in the book fully described, although structures encountered at each stage of its life history were described in detail and elaborately figured. Part of these figures show internal evidence of being drawings from actual preparations; others are obviously schematic or diagrammatic and present Brefeld's interpretation of his observations. The method of conidium formation was evidently not understood, but the general structure of the penicillus was beautifully developed, and the formation of perithecia as hard sclerotium-like masses of pseudoparenchyma followed by the slow development of ascogenous central areas was figured. Brefeld's work was done with the cruder methods of culture which preceded the rise of bacteriology with its provision for protecting cultures against contamination, hence it is not surprising if the various habit drawings presented suggest the probability that more than one species was involved in his series of cultures. At least one of his figures, Plate VIII, fig. 54, representing a coremium from a rotting pear, apparently represents some strain of *P. expansum*.

Before pure culture methods became well established, the custom of regarding a series of molds as polymorphic expressions of some single organism is well illustrated by Hallier's discussion and plate of his *Lyssophyton suspectum* (Zeitschrift f. Parasitenkunde 2: 72, Plate II, 1870) in which one figure out of twelve shows a *Penicillium*.

The use of the name *P. glaucum* for the apple rot organism was continued in Sopp's monograph (Sopp, p. 48) and in Wehmer's Beiträge. Both of them were students in Brefeld's laboratory, hence the continuity of this usage is evidence of understanding among them that this organism should be regarded as *P. glaucum*. Among the numerous strains of *P. expansum* which we have had in culture there has been no structure suggestive of sclerotia or perithecia during a period of more than twenty years within which time there have always been several to many members of this group in our collection.

Wehmer, in 1893, published his studies of the *Penicillia* occurring upon rotting fruit. He figured and described the destructive effects of *P.*

*expansum* on apples, pears and grapes, but under the name *P. glaucum*. The olive colored rot of oranges already described by Saccardo as *P. digitatum* in 1880 and distributed in Mycotheca Italici was called *P. olivaceum* and the soft rot organism with blue-green colors was correctly regarded as new and named *P. italicum*. In the same year he published his study of citric acid-forming molds to which he gave the name *Citromyces*, and his study of ascospore formation in *P. luterum* Zukal. Wehmer has given more attention to the biochemical and technical activities of molds than to their classification, although he prepared the studies of this group in the second edition of Lafar's Technische Mykologie.

Sopp beginning his studies of molds with Brefeld, records that he recognized as early as 1890 that the name *P. glaucum*, in addition to designating the apple rot organism, was being used to cover more than a single species (Elfving records the same conclusion in 1895). In 1898, Sopp separated certain of these forms found active in cheese ripening as *P. aromaticum*. Unfortunately he gave entirely inadequate descriptions of these organisms, apparently depending for identification primarily upon their presence upon particular varieties of cheese. Even in his monograph in 1912 his usage remains too indefinite to be verified. His monograph brings together a mass of descriptive matter and many figures covering some sixty species. In spite of the mass of data presented however, very few of his species have been certainly identified by any other worker. His early papers were signed Olav Johan-Olsen.

Dierckx published his "Essai" in 1901, in which he sketched his method of work and proposed a series of twenty-five names with descriptions so brief and inadequate that no one working with the published material succeeded in identifying any of his new species. He appears to have left in the laboratory at Louvain, colored plates, descriptive material and sufficient of his cultures to enable Biourge to reidentify and redescribe twenty-two of these forms in 1923. No one of Dierckx's forms was positively identified with any previously described species, although some names were used with an interrogation point, and two of them apparently correctly. As noted by Saccardo in the Sylloge, the new names proposed were not supported by descriptions or figures which would identify them. Biourge in his monograph bases his recognition and perpetuation of some of Dierckx's specific names upon the accessory material left by Dierckx. With reference to certain of these forms, which were adequately described by others between 1901 (Dierckx's paper) and 1923 (the date of Biourge's monograph), we are not disposed



to recognize the validity of the incomplete and unrecognizable descriptions of Dierckx's paper as taking precedence of adequate descriptions by others.

Biourge also reproduces under Dierckx's numbers a series of descriptions of unnamed forms assigned to *Biverticillium* (*Biverticillata-symmetrica*). From his failure to complete the descriptions and name the species it may be safely inferred that the cultures were lost and that Biourge himself had failed to fix upon strains which could safely be identified by the descriptions. His comment that these numbered forms are better described than many of the old species of the genus may be true, but since it is equally true that no one has recognized these organisms of Dierckx, it seems better to disregard them all in so far as the date of Dierckx's paper is concerned. Such forms as were finally properly identified and described in Biourge's paper but attributed to Dierckx by Biourge are regarded in this book as published by Biourge although the reference to Dierckx's paper is given also.

Thom began to study *Penicillia* in connection with cheese ripening in 1904, published "Fungi in Cheese Ripening" in 1906, his "Cultural Studies" in 1910, his group conception of the classification of the *Penicillia* in 1915. In these papers the necessity of comparative culture upon standardized media was stressed in contrast to the search for optimum conditions of culture, organism by organism. The present work summarizes a continuous observation of species of *Penicillium* in many thousands of cultures over twenty-five years.

Weidemann in Kiel (1907) and Westling in Stockholm (1911) used careful cultural methods in describing their series of green *Penicillia*. Neither worker had a large enough collection nor followed the study long enough to determine group relations involved.

Dangeard (1907) included "*P. glaucum*" and discussed "*P. vermiculatum* n. sp." in his extensive study of peritheciium formation. Unfortunately in his study of *P. glaucum* he included among figures (Plate XV, figs. 5 and 6) of the conidial apparatus, forms which clearly belong to some species of *Scopulariopsis*, (*P. brevicaulis* and allies). His cultures of "*P. glaucum*" were therefore impure and little attention can be paid to the findings. In *P. vermiculatum* he described some member of the *P. luteum* group with uninucleate cells, conidia 3 by 2 $\mu$  and rough, asci 4 to 6 spored, elliptical and (Saccardo Syll., 22: 1278) spinulose, but left the description so incomplete that recognition is questionable. Derx without giving any evidence decided that the form designated *P. luteum* by Thom (1910) and sent to Derx as Thom No. 11 was *P. ver-*

*miculatum*. Since the conidia do not agree with Dangeard's description and the asci are 8-spored, the conclusion appears to us doubtful until Derx supplies his evidence. The general morphology and cytology of perithecium and ascospore formation may have been correctly given in Dangeard's figures and hold for a whole series of related organisms without establishing identity.

Bainier continuing his "Mycothèque de l'École de Pharmacie," at Paris began publishing his series of species of *Penicillium* in 1905 and continued partly separately, partly with Sartory until 1914. In this series of species many forms were described and figured with meticulous care, hence some of them are readily identified.

Efforts were made to maintain all of these forms in culture at l'École de Pharmacie (see Chapter III). Bainier separated the form described later (1910) by Thom as *P. divaricatum* and made it the type of his genus *Paecilomyces* (1907), but we have found no record of the recognition of this genus from Bainier's description and figures. When we obtained his culture (no. 4640.436) and studied it in connection with his description, identification was possible. He also designated *P. brevicaulis* Saccardo with its numerous allied forms as the basis of a new genus, *Scopulariopsis* (1907). The separation of both of these groups from *Penicillium* can be readily justified on the ground of lack of essential relationship, but the inadequacy of the descriptions furnished as a basis of recognition is well illustrated in the case of *Paecilomyces*. Bainier adopted Wehmer's genus *Citromyces* and described his monoverticillate strains as species under that name. He failed, however, to indicate relationships among his *Penicillia* so that in spite of every effort we have been able to make, a number of his species remain very uncertain.

Biourge under the stimulus of Carnoy began to study *Penicillia* in 1897 in connection with his other undertakings. After Dierckx had published his "Essai" and abandoned the completion of his monograph, Biourge took up the task in earnest. He gave a brief survey of this work in a conference at Louvain in 1916, which was published as a pamphlet in 1920, followed by his monograph in 1923. In this work, Biourge followed upon the culture lines already proposed by Dierckx, broadened his work to cover most of the studies made by Thom and gave a brief Latin diagnosis for some 125 species, followed by culture notes in French, and supplemented by line drawings of the conidiophore, penicillus and conidia, and colored plates showing the colors and color changes of his colonies.

Unfortunately Biourge's microscopic work was almost entirely done

from colonies removed from cultures preserved in alcohol. His diagrammatic sketches equally lack all touch with the growing colony. He lost therefore the habit concept with its large contribution to our knowledge of the organism as it grows in the petri dish. His measurements as far as significant appear to have been carefully made but the numbers of sterigmata, metulae, branches, etc., specified, while correct to his drawings, do not tally with studies of living colonies of his own strains as grown in our media.

Biourge's conception of the genus in the broadest sense calls for the inclusion not only of *Coremium*, *Citromyces* and of more or less related series like *Scopulariopsis*, *Gliocladium*, and *Paecilomyces*, but part or all of *Stysanus*, *Isaria*, *Spicaria*, *Monilia*, *Oospora*, *Briarea* and other genera in which figures or descriptions offer suggestions of a penicillate conidial apparatus. He therefore included a "list onomastique" (Monogr., pp. 100-106) in which the species selected are given as transferred to *Penicillium* with the alterations of specific names indicated in such transfer. For the sake of completeness of reference all of these names are included in the index but those for which no justification is seen except adherence to the bibliographic transfer of a group from genus to genus are indicated as such by a note but are given no other reference.

Zaleski (1927) described thirty-five species and one variety of *Penicillium* from the soil under the forests of Poland. His macroscopic observations were recorded from cultures grown in petri dishes on neutral Raulin's solution with 10 per cent gelatin. His microscopic data were obtained from colonies upon the same basal substratum but with agar (1.5 to 1.7 per cent) instead of gelatin. The cultures were incubated at 17 to 19°C., and observed at 6, 10, 12, to 18, 24 (or 26) and 30 days. His Latin descriptions repeat themselves frequently and go into excessive detail of the buckling and wrinkling of colonies upon gelatin as the substratum becomes liquefied. In our experience, wrinkling and buckling in a liquefied plate is a very unstable character. Nevertheless new taxonomic sections were based upon the shape and arrangement of these wrinkles. His cultures and notes were sent to Biourge whose comments are given for certain species but whose opinions were not always followed. Zaleski placed much emphasis upon macroscopic colony appearances in gelatin plates but failed to take account of some important group differences. Apparently he confined his study to these soil organisms hence had no comparative back-ground in acquaintance with the many cosmopolitan saprophytes belonging to the group.

No microscopical observations upon the undisturbed colony in the

petri dish were given. Measurements and details of structure, and arrangements in fruiting hyphae or conidiophores were made from colonies 8 to 12 days old, after the material selected had been washed with water several times and carefully teased. This method follows Biourge in the complete destruction of much of the most useful information as to the structure and appearance of these conidium-producing organs as they are developed in the colony.

The whole question of size, arrangements, measurements and numbers of parts was discussed by Zaleski in his third section (p. 517) where he concluded that differences in size of conidia mean little in *Penicillium*. Similarly he believed that the presence of a connective (isthmus, bridge, disjunctor) between the conidia has little systematic value; also that the measurements of sterigmata show too wide variation to make small differences significant. In shape he reported sterigmata slender (schlank), cylindrical, flask shaped, or fusiform, some curved; the conidium bearing tube as long, or short and heavy. Zaleski seems to have been convinced that the general shape is characteristic, but that the number of sterigmata to the verticil is not a satisfactory character to separate species (his figures are far too small), although the variations within the species are significant. As to metulae the exact measurements are not deemed very characteristic but the general size and shape of the metulae hold good.

Zaleski accepted the subgenus, section, subsection, and series divisions as given in Biourge's monograph and fitted his species into the scheme as far as possible. In Biourge's section *Biverticillium*, he proposed four new subsections among which he divided the ten species assigned to *Biverticillium*. Among the cultures received as his types only one belongs in the symmetrically biverticillate division (Chapter XX). The remainder belong in the monoverticillate, and in the asymmetrical and divaricate sections in which biverticillate penicilli are fairly regularly found mixed with the monoverticillate type.

Recently Scaramella, Arnaudi, and Ullscheck have each given rather detailed cultural reactions for a series of species or strains but in each case the morphological data have been too meager for identification of cultures. The physiological value of their researches is not questioned especially where general principles were obtainable, but, since many reactions or tolerances, especially quantitative reactions, are characteristic of species or even of strains, identification which would make repetition more easily accomplished would have added greatly to their value.

## CHAPTER III

### HERBARIA, EXSICCATI, CULTURE COLLECTIONS

#### EXSICCATI

Herbarium specimens in the *Penicillium* group have identification value in dealing only with an occasional species. Specimens in Saccardo's *Mycotheca Italica* (no. 986) labeled *P. digitatum* yielded recognizable conidia which satisfactorily verified the identification. Ravenel's collection labeled *P. roseum* no. 1179 in DeThümen's *Mycotheca Universalis* was identifiable with organisms in culture. Lately we have checked *P. repens* of Cooke and Ellis against a culture from a chestnut and found the result satisfactory. On the whole the old collections are hopeless as a means of determining what organisms the describers had. The exsiccati in the Pathological Collections of the United States Department of Agriculture, the herbarium of the New York Botanical Garden, and that of Harvard University, have been examined carefully. The collections at Kew and Berlin were seen but offered little encouragement for intensive study. No attempt will be made to cite specimens in these collections in connection with the description of species.

The mycelium and conidial fructifications of most species of *Penicillium* are exceedingly fragile, especially after the colonies have become old and dry. When exsiccati of natural substrata carrying colonies of *Penicillium* are examined, the mass usually breaks up completely in the mounts used. Even the conidia in some species have a shriveled or crushed appearance. Using a proper treatment the conidia will swell to resume their original outlines, hence make possible determinations of size and shape. But penicilli of most species separate into their single cells. Colonies grown in laboratory media, then transferred to the bottom of flat paper boxes (pill boxes) can often be made to adhere to the bottom of the box and dry to give a good habit picture of the colony. Such specimens are valuable as illustrating the general appearance of the species, but no more useful for microscopic studies than exsiccati from natural sources.

If space permits, cultures in the original tubes or petri dishes may be dried and placed in the herbarium. Such specimens offer more favorable material for identification than any except the occasional form which produces special structures upon particular natural substrata. Maintenance of type cultures in this form is exceedingly desirable.

## TYPES

By the method of types in general botanical taxonomy, the description of a species must apply to a particular specimen, marked as type and deposited in some accessible collection for reference. Application of the rule to species of *Penicillium*, is very desirable even though it gives only limited satisfaction. Saccardo's material of *P. digitatum* consisting of a piece of moldy orange peel proved convincing. Jensen's *P. terrestre* made reference to a morphological series moderately safe but scarcely identified a particular culture. Many of the species can be grouped but few of them can be separated from related species in the group by such material. Such material should be supplemented by maintaining type cultures by transfer from time to time to keep vigor and viability. Theoretically, this is possible. Practically, as long as the cultures remain in critical hands and are critically examined from time to time, the results are fairly dependable for some species. In other species transfers upon a routine substratum may appear entirely satisfactory for a time followed by loss of vigor and change of reaction or, at times, such change in colony appearance as to lose the characteristic picture set forth in the description. This is often due to contamination or replacement by another species but it has been seen often enough under conditions rendering that interpretation improbable to lead to the belief that some species can only be kept for long periods by special study of their requirements. Unfortunately numerous types slipped away before the necessity of special methods was realized, hence some are gone from our own collection and many from the collections of others obtained by exchange.

## COLLECTIONS OF LIVING CULTURES

In recent years, systematic collection of living organisms to be maintained in culture has become an important adjunct to the preservation of material, which has been newly described or has formed the basis of specific investigations. Two attitudes of mind are legitimate in the making and preservation of such culture collections: (1) scrupulous care in obtaining a culture directly from a describer and its perpetuation as received; (2) scrutiny of every culture received for purity followed by identification by comparison with published descriptions. The one proposes to obtain a culture vouched for by a particular worker and pass it along with the voucher attached. The other seeks to handle only organisms truly representing species, varieties and strains described or connected with processes or products which render the preservation of

the organism academically or technically important. The monographer must restrict his own work to the second type of culture without losing sight of the fact that many cultures of the other type will be received and must be discreetly handled. Collections of *Penicillia* have been made in various countries over a period of about thirty years. Only a few laboratories have made extensive collections of cultures in this group.

Brefeld does not appear to have maintained a collection of this kind although he was called upon and personally asked for material in 1905. Johan-Olson Sopp appears to have kept his organisms for several years but did not distribute to others for study.

Wehmer, when visited in 1905, maintained no collection but gave freely of his experience in examining fresh samples of decaying fruits and vegetables and in examining the cultures carried to his laboratory. Further, he has from time to time contributed cultures with his nomenclature to our collection.

Professor Thaxter at Harvard has contributed occasional cultures from his personal collection.

Bainier at Paris maintained part at least of his collection until his death and the laboratory continued to transfer his species for several years. The collection was examined by daFonseca, who sent to us transfers of all *Penicillia* he was able to find. This collection was exceedingly valuable, but rigid scrutiny quickly showed that the original organism had been entirely replaced in some cases by a very different species while the names on other tubes had been interchanged. It seemed fair to believe, however, that any organism found in that collection had probably passed through Bainier's hands. The entire collection was, therefore, studied to locate as many of Bainier's types as possible. Several forms were recovered in this way which were not correctly represented by the labels but confirmed us in our interpretation of his descriptions.

Among these cultures *Paecilomyces varioti* proved to be identical with *P. divaricatum* Thom, and *P. caseicolum* with *P. camemberti* var. *rogeri* Thom. *P. urticae* fixed the identity of our 2694 received from Miss Dale as isolated from English soil. *P. elongatum* was correctly labeled but had also replaced several of Bainier's original forms. *P. herquei* established the identification of some of our own cultures. Scrutiny of the remainder showed that few if any of them would satisfy the descriptions given by Bainier. Upon the assumption that organisms coming from the laboratory in which Bainier worked, might have been members of the original collection each of them was checked against

each of Bainier's species. In this way we found no. 4640.451 labeled *P. patulum* but actually corresponding to *P. paxilli* which in turn antedates *P. stoloniferum* of Thom. *P. patulum* was found in no. 4640.454 which came to us as *P. puberulum*.

• Such an experience is fair warning against the acceptance of labeled tubes as representing species and furnishing the proper basis for biochemical work or subsequent identifications unless the organism conforms to the published descriptions and figures of the species claimed. This was repeated when we received from two biochemical laboratories in Germany, tubes labeled *P. variabile* Wehmer but containing *P. chrysogenum* whereas Wehmer's own culture forwarded at the time he published his description was closely related to *P. expansum* and was so recognized by Biourge in his monograph.

Westling upon the completion of his paper sent to Thom transfers of all the species remaining in his collection. Several of his cultures as received, however, proved unsatisfactory, hence there remains much uncertainty as to the type back of certain of his species. Unfortunately he found it impossible to continue even the identification of cultures sent to him.

#### *The Centraalbureau*

Dr. Johanna Westerdijk, in charge of the Centraalbureau voor Schimmelculturen, Baarn, Holland, has freely exchanged with us all the *Penicillia* which have been deposited with her. In 1928, she passed over to us the whole Zaleski collection. These cultures show evidence of careful handling to maintain the organisms in the condition in which they were received by her.

#### *Kral-Pribram*

In 1924, we purchased from Pribram at Vienna all the *Penicillia* of his collection. Some of these cultures were true to type and useful additions to our collection, but the large majority of the original molds had been entirely replaced by contaminations.

#### *Biourge*

• After the publication of his monograph, Professor Biourge prepared transfers of those of his type cultures which remained viable. These have been studied in connection with the text of his monograph and our direct cultural experience is indicated by notes in the discussion of his species.



*Thom and Church collection*

Our collection began at the Storrs Agricultural Experiment Station in 1904 when Thom began to collect the molds found in connection with the cheese industry. The collection was first broadened to include the organisms found upon forage and feeding stuffs, again to include the molds isolated by Professor Esten and Miss Mason in their studies of soil microbiology. In 1914 the project was moved to the Bureau of Chemistry and again broadened to cover the organisms encountered in the microbiology of foods in general. From time to time workers in the field of human and animal pathology, notably Dr. F. Schmitter, have contributed their collections. Collections including few to many species for study and identification have been contributed by the following investigators: Chung from Chinese sources; Linder from British Guiana; Brierly from Rothamsted Experiment Station; C. G. Hansford from Kingston, Jamaica; J. R. Johnston from Porto Rico; Putterill from Cape Town, South Africa; Van der Bijl from Pretoria; Saito from Dairen, Manchuria; Hanzawa from Sapporo; Da Fonseca from Rio de Janeiro; the laboratories of forest pathology at Madison, Wisconsin and in Washington; Werkenthin and Lewis from Texas (soil organisms); Wilson and Waksman from the laboratory of the New Jersey Experiment Station; Fawcett from Riverside, California; C. E. Burnside from his studies in the pathology of the honey bee at Ann Arbor, Michigan; Mrs. M. N. Kidd's collection of apple storage fungi from London; Schwarz from the Gold Coast, West Africa. Many single cultures from widely separate sources have added interesting and helpful material.

*British Cotton Industry Research Association*

The British textile workers have established a mold collection at the Shirley Institute, Didsburg, Manchester, to serve their own needs. As a great courtesy, we have received transfers of the species of *Penicillium* present in their collection.

*Nobel's Explosions Company*

The Biochemical Laboratory of Nobel's Explosions Company has conducted research upon molds as agents of fermentation over a period of years. By courtesy of Dr. H. Raistrick and Dr. J. H. Birkinshaw, their whole series of species of *Penicillium* have been submitted to us for checking into this study of the genus.

*The American Type Culture Collection*

The collection of molds deposited with the American Type Culture Collection with headquarters at the McCormick Institute at Chicago University was brought together and maintained for several years by Miss Church and is now maintained in the Bureau of Plant Industry of the United States Department of Agriculture at Washington. We have made efforts to put into this collection actual type forms of the *Penicillia* which have been described by us or received as exchanges. Material not obtained from the describer is listed with reference to the worker contributing the culture and its identification.

This collection proposes to take over and maintain the species of the iousvar American collections deemed important either for research or technical purposes, thus eventually reducing the necessity of private collections to the species actually under investigation at any particular period.

## CHAPTER IV

### GENERIC CONSIDERATIONS AND USAGES

A hyphomycete genus such as *Penicillium*, based upon general resemblances in conidium production, is an aggregation of species with a common type of asexual fruiting, but not necessarily closely related in their so-called perfect form. Frequently closer scrutiny of the method of conidium formation points out lines of real relationship which are subsequently confirmed when the so-called "perfect" stage is discovered and described. So many species of diverse appearance have been described as *Penicillia*, that some orderly scheme of grouping must be found to facilitate identification before there can be hope of any consistency in the usage of various workers.

Many partial schemes have been devised for this purpose. These have mostly consisted of the separation of a species or a group of species from the whole aggregate with the proposal of a new generic name. Such a generic name if based upon a full description of life history including the ascosporic form presents a step toward permanent nomenclature. A generic name based solely upon differences in detail of conidial apparatus can be useful, and hence justified, if it separates a group so evidently genetically related that the separation facilitates the identification and study of its various species. If the proposed genus is an arbitrary grouping of forms upon a single not very fundamental character, it may merely complicate the problem of nomenclature without compensating advantages.

### SYNONYMY

All of these various proposals as to generic classification are reflected in the nomenclature of the *Penicillia*. Numerous bibliographic changes are encountered in the literature in which an author accepting or rejecting a specific generic change has applied that change to a series of species with or without evidence of critical acquaintance with the organisms themselves. We thus find that Biourge in his monograph has listed as changes to *Penicillium* a long series of published species described as *Isaria*, *Verticillium*, *Coremium*, *Spicaria*, *Gliocladium*, *Oospora*, *Citromyces*, *Paecilomyces*, *Trichurus*, *Scopulariopsis*. While some of these forms undoubtedly belong in the great group, many of these changes

were merely bibliographic adherence to a taxonomic decision resulting in listing among the species of *Penicillium* a great many forms which do not belong there.

The taxonomic sections of this book record our efforts to follow all of these changes in the assignment of species to the various genera involved, and to give as fair value as possible to these various proposals. It is quite probable that some species have escaped our notice as well as some of the many bibliographic changes recorded. It is, therefore, necessary to discuss a series of these generic changes separately, present the diagnostic characters used in them and to estimate their value. Most of them at best must be disregarded as genera although some of them remain as subgroups useful in the separation of members of a great aggregate, but not sufficiently fundamental to warrant their continuation as genera. These genera as discussed are presented in chronological order.

### *Coremium*

*Coremium* Link Obs., p. 19, 1809 (compare Chapter XIX, no. 350).

Link described *C. glaucum* on p. 19 after describing *P. expansum* on p. 17 of his "Observationes." Comparative study of decaying fruit has lead us to believe that *C. glaucum* was the coarsely coremiform development we now know to occur from the growth of *P. expansum* upon such fruits as apples in storage. Link's generic description follows:

"*Coremium*—Stroma capitatum, capitulo et stipite tectis floccis penicillatis. Sporidia floccis inspersa." [Note: Genus sometimes referred to *Stilbum*.] "at capitulum floccis spondiferis tectum est. Contextus tam stipitis quam capituli floccosus viditur. Flocci basi simplices in apice penicillati, tenerrimi. Sporidia minuta globosa. Hujus loci viditur *Monilia penicillus* Pers."

The name has continued to be loosely applied to coremiform fruiting masses wherever found so that several species are probably included in such aggregates as *C. vulgare*, Corda, *C. citrinum*, *C. candidum* Nees, and some of these as indicated in different numbers were in all probability species which we list here as *Penicillia*. No attempt has been made to include all species described under the genus.

Until someone restudies the whole *Stilbaceus* group of fungi and gives us a real classification, we may continue to use *Coremium* as a means of referring to some of these forms.

*Eupenicillium*

*Eupenicillium* Ludwig. Lehrb. niederen Krypt. Stuttgart, 1892, pp. 263-265.

The earliest of these proposals for separation was Ludwig's *Eupenicillium*, which merely accepted Brefeld's discussion of *P. glaucum* and made it the basis of an Ascomycete genus. Ludwig's discussion is headed, "Der gemeine Pinselschimmel, "*Eupenicillium crustaceum* (L.) Fr. (*P. glaucum*)" and consists of an abstract of Brefeld's discussion of *P. glaucum* with the generic name changed to *Eupenicillium*. Ludwig offered no real contribution to our knowledge of the genus and his proposals have been justly ignored by most workers.

*Citromyces*

*Citromyces* Wehmer, Beitr. z. Kennt. einh. Pilze I, pp. 1-92, Tafel I and II. Hannover and Leipzig, 1893.

Discussed by Westling, R. Arkiv. för Bot. 2: no. 1, p. 41, 1911; by Sopp, Monog. p. 184, 1912; by Thom, C., U. S. Dept. of Agr., Bur. Anim. Ind. Bul. 118, p. 23, 1910, and Biourge Monogr., La Cellule 33, fasc. 1, p. 32, 265, 331, 1923. (Compare Chapter XII.)

Wehmer proposed to separate into the genus *Citromyces*, penicillate species with conidiophores originating as branches of vegetative hyphae and not differing from such hyphae in origin or structure except in the development of a vesicle-like swelling at the apex upon which a single verticil of sterigmata is produced. The sterigmata do not differ from those of the typical *Penicillia*, and produce unbranched chains of conidia. No perithecia were described, but aggregations of hyphae suggestive of some form of fruit body were mentioned in the description of *C. pfefferianus*. The forms under observation by Wehmer produced citric acid by the fermentation of sugar solution, hence the name. All efforts to obtain the exact strains used in describing Wehmer's two species have failed, although the general morphology represented is readily found in many forms in our collection. It is probably logical to use these names to designate sections or series in the *Monoverticillata*.

Dierckx (Soc. Scientifique Bruxelles, 25: 83-89, 1901) uses the name *Aspergilloides* for the *Monoverticillate* section of the *Penicillia*, and Biourge in his monograph (La Cellule, 33: fasc. 1, 1923) uses the term *Aspergilloides* in his analysis on p. 32, but on p. 265 puts his own term *Monoverticillium* (from his 1920 paper, Bul. Ass. Anc. El. Ec. Brass.

Univ. Louvain, no. 3) first, follows it with *Aspergilloides* in bold faced type, and in the table, p. 331, puts *Monoverticillium* also in bold faced type.

Wehmer in a recent letter to Professor Raistrick adheres to his separation of *Citromyces* but does not undertake to identify species other than in the most general group terms. We know, therefore, the general appearance of the original species but no one knows particular strains as Wehmer's types. Citric acid production has since been shown to be a very general biochemic activity among several groups of molds rather than a particular characteristic of these species. The arguments for the name are not very convincing.

Species of *Citromyces* have been described by Sopp, Bainier, Bainier and Sartory, Mazé and Perrier, Pollacci, and mentioned or discussed by many others in connection especially with biochemical or technological studies. No one, however, can collect a large number of these organisms without finding that the separating marks believed adequate at first, actually fail since every gradation from the conidial apparatus of *Citromyces* to *Penicillium* may be found upon the same mycelium in some species, and that forms obviously nearly related would part of them fall in one genus, part in the other. We follow Biourge in dropping the generic name.

### *Microaspergillus*

Biourge included in his monograph under species names as *Penicillium*, a number of organisms figured with monoverticillate penicilli, which when he finally sent them to us in 1928, had been relabeled as species of *Microaspergillus*. In every case examined these species had the typical "footcell" of the *Aspergilli* (see Thom and Church p. 15) which in no case appeared in his previous drawings. On this basis alone, these organisms would be correctly removed from *Penicillium*. With the evidence before us that Biourge is preparing to publish his changes in nomenclature these species have been reported merely among the *species of other genera described as Penicillia* (Chapter XXVI). Evidently Biourge has taken up the name *Microaspergillus* which already appears in the literature (Wehmer) and has made it include a series of very small and delicate forms mostly with a single series of sterigmata in contrast to the coarser series such as the *A. glaucus*, *A. niger*, or *A. flavus* groups.

*Scopulariopsis*

*Scopulariopsis* Bainier, Bul. Soc. Mycol. France 23: 98. 1907. Synonym *Acaulium* Sopp. Type species *P. brevicaulis* Saccardo.

Bainier's discursive description presented valid observations together with characters that seem to us untenable, but study of a great many strains of the group lead to the conclusion that these organisms are not closely related to the usual types of *Penicillia*, hence may properly be segregated. Since *Scopulariopsis* was published in 1907 and *Acaulium* Sopp in 1912, *Scopulariopsis* may be accepted and an emended description proposed.

*Scopulariopsis* Bainier emended Thom.

Colonies never green; conidial apparatus partly *Penicillium*-like partly in reduced and variant aggregations of sterigmata and branches or even single scattered sterigmata; sterigmata either *Penicillium*-like or more or less specialized, tapering gradually from a basal tubular section toward a conidium producing apex from which successive conidia are cut off by crosswalls.

Conidia initially more or less pointed at the apex and truncate at the base with a more or less thickened basal ring surrounding a basal germinal pore, with walls usually thickened and often variously marked or roughened.

They appear as agents of decomposition after the usual green *Penicillia* have ceased to be active; that is, in the later phases of decay processes.

*Acaulium*

*Acaulium* Sopp. Monogr. *Penicillium*. Videnskap. Sk. I. Mat.-Naturv. Kl. 1912, no. 11, p. 42. Type species. *Penicillium brevicaulis* Saccardo. Synonym for *Scopulariopsis* Bainier.

Sopp's description of the conidial form adds nothing to the well-known characters of the arsenic fungus *P. brevicaulis* Sacc. He reports for certain of his species perithecia which show well defined ostioles, but stops short of such adequate descriptions of asci and ascospores as would ensure recognition of his species by these characters.

*Paecilomyces*

*Paecilomyces* Bainier, Bul. Soc. Mycol. France 23; 26, Plate 7. 1907. Probable synonym *Corollium* Sopp; *Spicaria* sp.? Type species

*P. varioti* Bainier: synonyms *Penicillium divaricatum* Thom, *Eidamia catenulata* Horne and Williamson.

Genus described as related to *Penicillium* and *Aspergillus*, distinguished from these genera by its sterigmata which are short tubular or more or less enlarged, tapering into long conidium-producing tubes mostly curved or bent slightly from the axis of the sterigmata; sterigmata occur variously, partly in *Penicillium*-like verticils with conidium-bearing tubes and conidial chains divergent, partly variously arranged on short branchlets, or again occurring singly and laterally upon fertile hyphae, in terminal groups often approximating the appearance of a *Penicillium*; conidia elliptical; colonies never green.

Bainier failed to describe accessory structures which appear more or less commonly in all species, and very abundantly in certain species of this group. These are regarded as macrospores by Horne and Williamson and made the basis of transferring these species to *Eidamia* (see discussion, Chapter XXIII).

Gilman and Abbott assign the whole series we have put in *Paecilomyces* to *Spicaria*. Jensen citing Oudemans put into *Spicaria* one species *S. simplicissima* which we have removed and placed in *Penicillium*.

### *Dactylomyces*

*Dactylomyces* Sopp. Monogr., p. 33, 35, 36, 37. 1912. Probable synonym: *Thermoascus* Miehe. Type species: *D. thermophilus* Sopp. cit. above.

This is a monotypic genus with no sharp lines of differentiation in the description given. The sole basis of separation is comprized in the following items:

Conidiophores bear finger-like branches at the apex, each enlarging to a vesicle-like apex, suggestive of the basidia of *Tomentella* (*Clavaria*-form), these branches produce similar secondary verticils and such branching may be repeated three, four or more times, hence the name *Dactylomyces*; perithecium formation begins quickly so that the cycle from ascospore to ascospore requires only a few days.

Aside from the general resemblance of the perithecium to that of the *Aspergilli* and *Penicillia* and the rough similarities of the conidial apparatus, the true affinities of the organism studied by Sopp are difficult to determine. We have not seen it.



*Corollium*

*Corollium* Sopp. Monogr., pp. 98-99. 1912. See also Olav Johansen [Sopp] in *Pharmacia*, no. 22 and 23, 1904. Synonyms: *Paecilomyces* Bainier.

Sopp's generic description can scarcely be separated from that of his type species, *C. dermatophagum*.

*Corollium* is reported to be related to *Acaulium* most nearly, but connects with the true *Penicillia* through *P. olivaceum* and *P. italicum*; appears upon moist leather as clear brown to yellow green, close felted mycelium becoming powdery or mealy in age with conidial masses of the *Penicillium* type at the apex of fertile hyphae, but accompanied by abundant verticils of sterigmata broadly and irregularly distributed on the fertile hyphae.

The type species of this genus was found upon a pair of old boots in a military depot in Norway, from which it appears to have spread as a leather destroyer over the whole land (?). It grows readily upon a wide range of media and is thermophilic.

From Sopp's figures and descriptions this organism was probably one of the series for which we are accepting the generic name *Paecilomyces* of Bainier.

*Carpenteles*

*Carpenteles* Langeron. Compt. Rend. Soc. Biol. Paris, 87 (Année 74), pp. 343-345. 1922.

Langeron proposes *Carpenteles* as a new genus to include ascosporic *Penicillia* naming *P. glaucum* (Link) Brefeld as type. This proposal is unsupported by new investigations of the species, hence acceptance must be postponed until someone puts data enough back of it to make the genus useful.

## GENERA SOMETIMES INCLUDED

*Penicillium*-like conidial apparatus occurs in cultures of certain organisms whose relationship to other well-known forms is established. In ordinary laboratory media some of these species lack the characteristic structures of those genera as collected in the field. Others produce these structures slowly or tardily, or upon special media. In this way species of *Isaria*, *Stysanus*, and, perhaps, other *Stilbaceae* have been identified and occasionally described as species of *Penicillium*. There is a horizon-zone of uncertainty in the separation of *Gliocladium* and

Clonostachys, from the penicillate types often designated *P. roseum* Link. Brief discussion of these generic names is pertinent.

### *Aspergillopsis*

*Aspergillopsis* Sopp. Monogr., p. 201-202. 1912.

This generic name was proposed for *Penicillium*-like forms in which the stalk of the conidiophore ends in a globose or clavate apex producing shorter or longer pearshaped or clavate "sterigmata" (or metulae?) radiating in all directions; each "sterigma" or metula bears a verticil of 5 to 15 short, thick, sterigmata, producing conidial chains.

Sopp regarded this group as presenting transition forms toward *Penicillium* much more frequently than toward *Aspergillus*. He described one species *A. fumosus*.

Thom and Church<sup>1</sup> in discussing *Aspergillus insuetus* suggest that it may belong with Sopp's genus *Aspergillopsis* if that genus is to be maintained for such intermediate forms as resemble both genera. In any case, the genus would need more accurate delimitation before it could be used, hence unless someone can determine just what type Sopp was considering, it may be dropped.

### *Isaria*

*Isaria* Persoon. Disp., p. 41, 74. 1797.

Many years ago, Atkinson pointed out the occurrence of *Penicillium*-like fruits in cultures known to arise from species of *Isaria*. This was readily demonstrated from time to time when insects bearing coremia of the *Isaria* type were brought in and cultures were made from their conidia. Studies of these forms gave a general type of structure fairly characteristic. Occasionally one of these forms produces coremia in agar suggestive of the insect parasite. Forms definitely recognizable as *Isaria* are not included in this book, although a brief discussion of one of them with a figure showing its detailed structure may be useful as a guide in interpreting such forms as they occur.

### *Stysanus*

• *Stysanus*, in Sopp Monogr., pp. 78-79, 1912, is designated as a sub-genus of *Penicillium*. Biourge follows Sopp in considering these species to be related.

<sup>1</sup> The *Aspergilli*, p. 154. 1926.

Upon gelatine, agar, earth, sawdust, both of the species studied by Sopp, produce Penicillium-like conidiophores, sterigmata and conidia; Sopp argues that the fact that the separate conidiophores arise from or diverge from a column of hyphae or a coremium, or for a part of their course form such a column does not warrant separation from the genus any more than the coremia produced by it upon apples warrants the exclusion of *P. glaucum* (*P. expansum*) from Penicillium.

Sopp especially notes similarities of *Acaulium fulvum* and *A. violaceum* to *Stysanus stemonitis* and predicts that *Stysanus* as a genus will eventually disappear in the Penicillium group. The real relationships based upon ascospore formation may be expected to take variant types which are now placed arbitrarily.

In our culture experience, species of *Stysanus* have dark almost black cell walls and differ so markedly in general aspect as to suggest relationships widely divergent from the usual types of Penicillium. The form-genus *Stysanus* can well be maintained and these species excluded from Penicillium. Dematiaceous species with brown to black cell walls as found on natural substrata occasionally appear with walls colorless in laboratory media during the first few days of growth. It is entirely possible that such species may be included among the Penicillia unless the observations are carried farther than we sometimes do.

Sopp includes in his monograph of Penicillium a description of *S. stemonites* Sopp which had been previously described by others and the description of *S. thyrsoideus* as new. Both of these diagnoses are in terms which show that the organisms in hand were not Penicillia in the sense used here. Biourge also listed and figured *S. stemonites* among the species studied as Penicillium; he contented himself with prophesying ultimate inclusion in the genus. The species of *Stysanus* have been excluded.

#### *Clonostachys*

*Clonostachys* Corda, Prachtflora, p. 31, Tafel XV. 1839. Type species *C. araucaria* Corda.

Corda's diagnosis included: Mycelium creeping, continuous (?); stalks erect, simple, continuous, verticillately branched above; each branch bearing 2 or more superposed verticils of 4 sterigmata each at successive nodes; sterigmata (Lat. ramuli) subulate with apex subcapitate, bearing spores spirally, forming a kind of spike; spores unicellular, with walls hyaline and contents curved around a central globule.

Further description in the light of more recent studies of spore forma-

tion has not been found. Corda's figure and description have been interpreted as covering certain fruiting forms occasionally encountered. We have had several cultures showing a penicillus in which the column of conidia assumed the appearance indicated by Corda's figures. The branching system found lacks the definite verticils of 4 sterigmata at each node prescribed by Corda, but reproduces the figure of *P. roseum* as given by Thom, 1910, which is essentially that of a *Gliocladium*. In fact, many observations suggest that these rosy or salmon forms which may show a penicillate condition at one stage, a *Gliocladium*-like fruit at another and again *Clonostachys* heads upon the same mycelium, should be restudied and some usage devised which will indicate the real relationships involved and reduce the other names to proper synonymy.

*Gliocladium*

*Gliocladium* Corda, *Icones Fungorum*, IV; 30-31, Taf. VII, fig. 92.

1840. Type species: *G. penicilloides* Corda, *Icones Fungorum*, IV: 30-31, Taf. VII, fig. 92. 1840.

Latin description repeated in *Icones*, V, p. 14, 1842, with a brief paragraph in German. See also Matruchot, *Rev. Gen. Bot.*, 7; 321, Pl. 16, 1895; and Bainier, *Bul. Soc. Mycol. France*, 23: 111-112, Pl. XV, 1907.

The essential characters given by Corda were: Conidiophores erect septate, penicillately branching above; branches and branchlets septate, appressed, forming a solitary gelatinous head; conidia unicellular borne upon the tips of branchlets and held together by mucilaginous substance in a dense mass.

*Gliocladium* was thus described as reproducing the growth habits, mycelium, conidiophores and conidial apparatus of *Penicillium* except that the conidia borne successively from the tips of sterigmata do not adhere in chains but become enveloped in mucilaginous drops which increase in size with the increased numbers of conidia, followed by the fusion of the masses upon adjacent sterigmata, then often the fusion of these mucilaginous masses with those from adjacent penicilli to produce large balls of conidia.

• Matruchot has described perithecia and ascospore formation in certain species, but the forms constantly encountered in culture are purely conidial. Comparative studies of structure in both conidial and ascosporic forms are necessary before *Gliocladium* and *Penicillium* or its ascosporic sections can be safely placed with reference to each other among the Ascomycetes.

*Lysipenicillium*

*Lysipenicillium* Brefeld. Unters. Gesamtgeb. der Myk., 14: 209–210, 1908; and 15, Taf. VII, fig. 1–7, 1912. Type species: *L. insigne*, was probably *P. insigne* Winter, but identity is not claimed.

Brefeld in describing *L. insigne* separates a species which is shown by his descriptions and figures to be a member of the *Gliocladium* group. He failed to give an adequate generic characterization, thus throwing upon one who chooses to accept the name the necessity of seeking the proper characterization in previous publications, none of which are cited.

Thaxter (1922) expressed the belief that *L. insigne* Brefeld was *Gliocladium penicilloides* Corda.

*Synpenicillium*

*Synpenicillium* Costantin, Bul. Soc. Mycol. France, 4; 62–68, Pl. XIV, figs. 10–17, 1888. Type species; *Synpenicillium album* Costantin. Synonym: *P. costantini* Bainier, Bul. Soc. Myc. France, 22, pl. 11, figs. 1–6, 1906. Synonym: *Coremium album* (Cost.) Sacc. and Trav. Change of name only in Syll. Fung., 19: 428, 1910; diagnosis Syll., 22: 1444, 1913. Incorrectly spelled: *Sympenicillium* Biourge Monogr. p. 216.

Costantin created his genus *Synpenicillium* for one species, *S. album*, specifying as the generic character the production of short conidiophores from fasciated (ropy) hyphae and as aggregates of several conidiophores arising together from fasciated hyphae.

Bainier regarding his species as identical with Costantin's, discarded the generic name and since *P. album* was preoccupied proposed the name *P. costantini* for this form. The name was again changed by Saccardo in the Sylloge to *Coremium*, but on inadequate grounds. Bainier's name is accepted here (see Chapter XXIII).

## CHAPTER V

### CULTURE OF PENICILLIA

#### SOURCES OF PENICILLIA

A few species of *Penicillium* are so closely associated with particular products or processes that presence of a *Penicillium* in a particular situation warrants presumption of an identity which is readily confirmed by microscopic examination. Most of these species will readily grow in other conditions and upon other substances and when so grown commonly differ so greatly in gross appearance as to fail to suggest their identity even to one familiar with them as rots. In such situations, the examiner may be (but usually is not) able to recognize his organism by microscopic examination alone.

#### CULTURE A NECESSITY

A still larger number of *Penicillia* have not been shown to bear any specific relation to particular processes and products. Such organisms appear as part of the complex population of stale or decaying organic matter, producing areas or tufts of white, blue, green, yellow or brownish mold, sandwiched in, overgrowing or overgrown by masses of *Mucor*, *Aspergillus*, *Fusarium*, *Cladosporium*, *Alternaria* and a multitude of less frequent organisms. The descriptions of *Penicillia* found in the literature up to the beginning of Wehmer's work, mostly represent the microscopic examination of preparations made from selected spots in such conglomerate mixtures. As early as 1890, Sopp reports that he found the current descriptions of *Penicillia* unsatisfactory, hence turned to culture as the best hope for a stable nomenclature.

The taxonomist must look to these natural substrata with their complex flora and fauna for his species of mold and render his practical service in the degree to which he correlates his findings from the study of the species isolated, with the conditions, or processes encountered in nature or in industry.

#### CULTURE AND CULTURE MEDIA

The development of artificial culture methods made possible the isolation and study of the habits of the individual mold. Along with

the academic interest in mold itself, the significance of some of these molds as the active agents of decay, fermentation, or ripening activity gave such studies an increased importance. Wehmer showed that two species of *Penicillium* were leading causes of decay in oranges and scarcely found elsewhere. Two other of his forms proved active enough in the production of citric acid by fermentation to warrant the establishment of factories. Sopp isolated the active agents in ripening certain cheeses highly esteemed for their flavor.

These developments in studying *Penicillium* merely paralleled studies of the biochemical activities of other microorganisms, which had already progressed much farther with *Aspergillus*, *Mucor*, yeasts and bacteria. The practical problems of combating, controlling and utilizing molds have now yielded enough results to point out lines of development toward much greater progress in the future.

For the culture of *Penicillia* the media already developed in the study of yeasts, *Aspergilli* and bacteria were first utilized. Most species of the group grow readily upon any culture medium used for bacteriological or mycological work. Their spores are so small and light as to float in the air and to be carried about by every breeze or motion. They become, therefore, the most abundant and persistent of "weeds" in the culture room.

Naturally for organisms growing so readily, students of *Penicillium* have usually employed formulae already in use in their laboratories except as particular purposes called for the development of specific combinations. Every kind of medium has, therefore, been used and work has been recorded upon many varieties of nutrient. Certain formulae have, however, been extensively used in the description of species of this group and these will be more or less fully discussed.

The solution proposed by Raulin in his study of the biochemical activity of *A. niger* has been widely used.

*Raulin's solution*

	<i>grams</i>
Water.....	1500.0
Cane sugar.....	70.0
Tartaric acid.....	4.0
Ammonium tartrate.....	4.0
Ammonium phosphate.....	0.6
Potassium carbonate.....	0.6
Magnesium carbonate.....	0.4
Ammonium sulphate.....	0.25
Zinc sulphate.....	0.07
Iron sulphate.....	0.07
Potassium silicate.....	0.07

Dierckx's neutral Raulin's fluid as given by Biourge, p. 43, follows:

1. Dissolve 0.40 gram carbonate of magnesium in a 100 cc. flask with 0.71 grams of tartaric acid.
2. In a liter flask with 800 to 900 grams of distilled water, dissolve saccharose 46.6 grams, ammonium nitrate 2.66 grams, ammonium phosphate 0.40 gram, potassium carbonate 0.40 gram, ammonium sulphate 0.16 gram, zinc sulphate 0.04 gram, iron sulphate 0.04 gram.
3. Add 66 to 67 cc. of the solution of magnesium tartrate (No. 1 above) and make up to 1000 cc. with water.

Biourge, on p. 37, notes that the purpose of this revision is to eliminate the free tartaric acid of Raulin's original formula by using only enough to dissolve the magnesium carbonate hence he appends in a note that he uses magnesium carbonate 0.27 gram, tartaric acid 0.40 gram, grinds them in a mortar with a few drops of water until dissolved, dilutes at once to large volume to stop crystallization. Biourge regards this as a good substratum when 10 per cent gelatin is added but as an indifferent or even poor nutrient when agar is used to produce a solid medium.

This substratum was used by Zaleski in describing his species but Zaleski modified the method of preparation as follows: (freely translated). The tartaric acid crystals were shaken in lukewarm distilled water and let stand until dissolved. The magnesium carbonate was then added and dissolved at once. The ammonium salts were dissolved in separate vessels with the water slightly warmed. The rest of the components were dissolved together in about 300 cc. of water. The gelatin or agar was dissolved, separately in about 500 cc. of water. After the gelatin or agar was fully dissolved, the other solutions were added while the mass was stirred. No precipitate should fall when the mixture is properly made. No filtration or clearing process was needed with high grade gelatin. The gelatin medium was sterilized in steam on the first and third days. Agar media were autoclaved. Layers of substratum 4 to 5 mm. deep were used in the petri dishes.

Biourge sometimes used 0.75 per cent of agar-agar and 5 per cent of gelatin with this solution to prepare a solid substratum for general culture work with saprophytic molds. Zaleski made his solid substratum by adding 1.5 to 1.7 per cent of agar-agar or 10 per cent of gelatin.

#### *Kauffman's formula*

Burnside gives (unpublished MS.) the following as a substratum favorable for molds but unfavorable for yeasts, in general use in the



mycological laboratories of the University of Michigan (attributed to Prof. C. H. Kauffman):

Maltose.....	5.0 grams
Magnesium sulphate.....	0.10 gram
Calcium nitrate.....	0.50 gram
Potassium dihydrogen phosphate.....	0.25 gram
Agar.....	15.0 grams
Water.....	1000.0 cc.

*Wort or beer wort*

Brewery wort or beer wort formed the basis of much of the media used by Wehmer, Sopp, Dierckx, Lindner and many other workers. Biourge insists that it be always regarded as the normal medium for *Penicillia*. Few of them specify more closely the characteristics of this liquid. Biourge alone offers some general limitations. After many unsatisfactory experiences with brewery wort and confronted with increasing difficulties in obtaining it in America, we followed Blakeslee in his study of the *Mucors* in substituting an artificial wort made with dried malt-extract. A formula for this purpose was developed in our laboratories and published by Reddish. Certain modifications of practice have since been made. As used at present it is prepared as follows:

Dissolve 100 grams of Difco dried malt extract in 1000 cc. of distilled water. Add 12 to 15 grams of agar-agar, varying the quantity of agar according to the jelly-strength of the lot to produce the texture desired. Cook in the autoclave for ten minutes at 15 pounds pressure. Adjust to pH 7 to 7.2 with normal sodium hydroxide. Filter through cotton using suction. Tube or bottle then sterilize about fifteen minutes at 10 to 12 pounds pressure. If the temperature (or pressure) used in final sterilization exceeds that of the cooking process, a fresh precipitate will be thrown down. The color of the product as finished is a shade of brown approaching cinnamon brown of Ridgway XV.15' when in thin layers, thick layers and masses give deeper shades.

Sopp and Biourge regard wort in some variety as the "normal" substratum for *Penicillia*. If this merely means a substratum in which almost any mold will grow, it is correct. On the other hand, it is chemically a very complex or "shotgun" mixture hence no analysis of biochemical activities can be based upon the reactions obtained; it is not uniform in successive lots or in different laboratories; the color of the wort itself masks color reactions produced by the fungi. Nevertheless, it is very useful for stock cultures especially for maintaining deli-

cate species over long periods and should be available in some form where *Penicillia* are to be handled.

*Wort according to Biourge.* Select a pale wort (from the brewery) before the addition of hops, autoclave for fifteen minutes at 115 to 120°C., filter in the boiling condition, distribute in tubes or flasks and sterilize fifteen minutes at 120°C. The density at 4.8° to 5.6°C. should be 12 to 14° Balling.

*Wort-gelatine-agar according to Biourge.* Biourge prepared his wort-gelatine-agar substratum by dissolving 1.5 per cent of agar-agar in the wort, using the autoclave at 120°C. for a half hour, then adding an equal quantity of wort containing 10 per cent of gelatine, and sterilizing the mixture at 110°C. for fifteen to twenty minutes. Biourge's Latin diagnoses appear to have been based upon cultures made with this substratum.

### *Hayduk*

Dierckx in one of his cultures of each species used a formula attributed to Hayduk. Biourge also reports the reactions of his species upon Hayduk's fluid. The formula follows:

	<i>grams</i>
Water.....	1000.0
Dipotassium phosphate ( $K_2HPO_4$ ).....	1.0
Magnesium sulphate (crystallized).....	0.32
Asparagine.....	0.80
Sucrose.....	80.0

To this he added 10 per cent of gelatine or for a gelatine and agar medium, agar-agar 0.75 per cent and gelatine 5 per cent.

### *Licorice sticks*

Bainier's descriptions lack any specification of the substratum upon which the colonies were grown for his diagnostic observations. Many references are made in his discussions to the use of licorice sticks for culture purposes. The cultures seen in his laboratory in 1905 and others received from Gueguen at the laboratory were grown upon licorice sticks in tubes, above a water holding constriction. Bainier's descriptions have been read many times and present only such essential data as to colony characters as could be made upon these licorice sticks. No reference is made to petri dish cultures. This limitation adds greatly to the difficulty of interpreting his descriptions. Cultures upon licorice sticks grow well, and maintain their vitality for a long time, but in our

hands have proved exceedingly unsatisfactory in the separation of organisms closely related in the various groups.

*Bean agar and potato agar*

Thom in his "Cultural Studies" used potato agar and bean agar with, and without sugar. The method of preparation follows:

*Bean agar.* The directions for making bean decoction were obtained from Mazé at the Pasteur Institute in Paris. Common white beans are heated in five volumes of water. Boiling is stopped just before the swelling of the cotyledons would rupture the seed coats. This gives a clear, slightly yellowish liquid which filters readily yet contains sufficient nutrients to grow many species normally. Agar may be added as desired. Since this decoction is poor in available carbon, the addition of sugar is often desirable for many species.

*Potato agar.* The potato agar was selected because of its use in many mycological laboratories. In this medium uniform composition can hardly be claimed. The following process has been used in this work: The potatoes are carefully washed, pared, and sliced, then slowly heated for about two hours in approximately two volumes of water. At the close of the heating the water is allowed to boil. The whole is then filtered through cloth, and commonly through cotton also, water being added to make up the losses of evaporation and filtering. To this is added 1 per cent of shredded agar. It is then heated for from twenty to thirty minutes in the autoclave to 120°C. or higher, when it may at once be put into tubes for use, or, if cloudy, it may be very quickly filtered through absorbent cotton, after which it should be quite clear. The uncertainties in the composition of this medium result from the differences in the potato extract itself and from the fact that the difficulties in filtering this extract take out a varying amount, which is replaced with water. Titration shows that this medium is nearly neutral in cases tested, to phenolphthalein; consequently it is used without neutralizing. Culture and study of the same species upon successive lots of this medium show that these differences in composition have little if any effect upon the morphology of the species studied.

*Corn meal agar*

The corn meal agar of the plant pathologist is in general represented by the formula given by Shear and Stevens

"To 50 grams of corn meal add 1 liter of water. Keep in a water bath for one hour at a temperature of 58°C., never over 60°C. Filter through paper, add 1.5

per cent of agar flour, steam for one and one-half hours, filter, and tube. Autoclave for fifteen minutes at 115°C. Corn-meal agar made by the above formula generally tests +3."

A stiffer corn meal agar may be made by using 100 grams of corn meal and 15 grams of agar to 1000 cc. of distilled water, and heating all ingredients in the Arnold for forty-five minutes or longer, if necessary, to dissolve the agar. This medium is then handled without filtration.

Several laboratories using corn meal agar use quite varied quantities of corn meal to the liter of distilled water, and report the final unadjusted product as varying from pH 5.8 to 6.5.

### *Prune gelatine*

Westling's descriptions were based upon an infusion of about 10 prunes to a liter of water. To this he added 15 per cent gelatine to produce a mass firm enough to be handled in petri dishes. Our own cultures with this medium were entirely satisfactory for general purposes, but we abandoned the formula because however uniform it might be with a particular lot of prunes and the technique of a particular laboratory, it can not be described accurately enough to insure uniformity when prepared in widely separated laboratories. As a basis for biochemical observation the complexity of the materials presented reduce its value in analysing activities too greatly to warrant its use. At best it has no advantage over the wort type of culture medium.

### *Ullscheck's agar ("Nährstoffagar" of Ullscheck's paper)*

After extensive studies Ullscheck reports the following as the optimum culture substratum for his species of *Penicillium*:

Water, distilled.....	1000 cc.
Saccharose.....	50 grams
KNO <sub>3</sub> .....	10 grams
K <sub>2</sub> HPO <sub>4</sub> .....	8 grams
MgSO <sub>4</sub> .....	5 grams
Agar-agar.....	30 grams

He prepares this by heating the sugar and salts in half of the water at 100°C., for thirty minutes, cooking the agar in the other half of the water at 120°C., for fifteen minutes, mixing the two and heating to 100°C., for thirty minutes on three successive days. The stock is then ready to be tubed or poured into plates for use. Its reaction is pH 5.4.

*Czapek's solution agar*

Our collaboration with chemists upon the study of the biochemical activities of molds very early pointed to the desirability of a basal culture solution whose components were readily available in the stock room of any chemical or mycological laboratory. For this purpose Dox selected the general combination of reagents attributed to Czapek. The selection was intended to present each element in a single form as nearly as possible, thus making the elimination or substitution of one or the other component definitely significant in metabolism.

Culture data upon *Penicillia* grown in this type of medium have been accumulating since about 1909. In general, it cannot be claimed to be an optimum solution for *Penicillia*, but with few exceptions all of the

TABLE 1  
*Czapek's solution agar*

	AS ORIGINALLY PROPOSED	MODIFICATION NOT AFFECTING PENICILLIA
Water.....	1000.0 cc.	1000.0 cc.
Sodium nitrate.....	3.0 grams	2.0 grams
Potassium phosphate ( $K_2HPO_4$ ).....	1.0 gram	1.0 gram
Magnesium sulphate.....	0.5 gram	0.5 gram
Potassium chlorid.....	0.5 gram	0.5 gram
Ferrous sulphate.....	0.01 gram	0.01 or trace
Sucrose.....	30.0 grams	30.0 grams
Agar agar.....	15.0 grams	By test of sample, 12 to 20 grams

species studied will grow and produce characteristic colonies upon such media. Failure to grow well, therefore, becomes a negative character of great usefulness. Transfer back to this combination from whatever series of cultures we have tried, has usually brought back the colony characters originally recorded. Such a medium gains a decided advantage to the describer from failure to produce the maximum amount of growth, so long as the growth obtained has well marked character and maintains the vigor of the strain of mold studied. Our own descriptive data as given in this book have been based upon cultures where the formula was used unless otherwise indicated (see table 1).

In making this culture medium the neutral potassium phosphate is preferred to the acid form of the salt, since with the neutral salt the final reaction of the medium is neutral or only very slightly acid. Tubes of this substratum tested with the quinhydrone potentiometer showed

pH 6.8 to 6.9. Czapek's solution agar is not offered as an optimum substratum for any particular species, but as a mixture approximately neutral in reaction, which is readily made in any laboratory in fairly uniform manner, and which permits moderately vigorous growth of nearly all saprophytic *Penicillia*. The quantities of mycelium and conidia produced by many forms in other media are much greater, but for comparative study, a moderate growth of the majority of the species is more useful than the great masses of mycelium and conidia which are readily obtained by using enriched substrata.

#### CULTURE MAKING

In preparing cultures of *Penicillia* favorable for study, two types of containers should be provided in abundance test tubes and petri dishes. Other containers may be adopted for special purposes.

##### *Petri dishes*

The standard petri dish about 10 cm. in diameter, with flat bottom and top well fitted, to reduce but not entirely prevent evaporation, is best of all apparatus we have tried for producing colonies for examination. Gelatine or agar media sterilized in measured quantities in tubes will theoretically produce an equal layer in each dish, hence subject every transfer to approximately the same nutrient conditions. Such containers are necessary for "dilution cultures," into which the inoculum carried on the tip of a sterilized wire or loop is transferred to the melted medium in the tube, thoroughly shaken or whirled, loop or pipette transfers made from the first to second, and second to third, or further tubes to secure the dilution required, then the contents poured into the petri dish and allowed to solidify. For general purposes the dilution method is commonly abandoned and the melted medium is poured into the sterile dish and allowed to solidify before inoculation. For this purpose, the measured tube of nutrient is commonly abandoned for bottles of media each large enough to produce the desired layer of solid substratum in eight to a dozen dishes.

Bases and covers, although they may be carefully fitted at first, usually get mixed in washing so that in actual practice the fitting of dishes and covers is by no means uniform. As a result, the rate of evaporation in a series of dishes will vary so greatly as to offset the inequality of the layers produced by pouring the media from bottles. Biourge, therefore, goes further and pours a series of such dishes with one side raised so that the layer of nutrient is reduced to a thin film at one edge and remains a

thick mass at the opposite edge. A colony spreading over such a surface becomes subjected to a series of conditions in each dish which permits an increased range of observations to be made during the development of each colony, but makes a correspondingly increased demand upon the experience and judgment of the observer in interpreting his findings. We have followed Biourge's method very commonly in using slanted plates.

After the nutrient is solid enough to handle, the inoculum is transferred with a sterile wire or loop to the desired spot upon the medium in the dish. For special purposes the exact location may be marked in advance with a wax pencil upon the bottom of the dish. Where avoidance of the scattering of spores during transfer is important, the mass of spores may be transferred first to a water-blank (tube of sterilized water) shaken thoroughly and a single drop carried by a loop to the desired position in the dish. For general study purposes the "water-blank" is usually omitted.

#### *Test tube cultures*

Parallel with petri dish cultures, tube cultures are practically essential in the handling of every *Penicillium*. Inoculation of such tubes is usually transfer by wire or loop of a selected mass of mycelium or spores to the medium in a fresh sterile container. In maintaining our own collection or studying specimens received, transfers to a series of test tubes are made, as a rule, before any other studies of the colony in a particular container are begun. This perpetuates as nearly as possible the exact contents of the previous culture tube or of the specimen received. Dilution or selection may ultimately show the original culture to be valueless, in which case the whole series can be easily destroyed, but, if the initial series of transfers is omitted, subsequent studies may be seriously interfered with by unavoidable contaminations or accidents of handling. A half dozen tube transfers into Czapek, wort, potato agar, potato plugs, corn meal agar or special media are quickly made and give a closer insight into the habits and appearances of a strain of mold than single cultures made and studied with the utmost care.

#### *Hollow slides, hanging drops*

Ullscheck based his description of morphology upon cultures grown upon minute quantities of his "Nährstoffagar" (Ullscheck's agar) in slides as manufactured for hanging drop studies. Such a culture chamber or depression was covered with a sterile cover glass held in place by

vaseline which was omitted along about one-fourth of the line of contact thus insuring ventilation while remaining tight enough to exclude contamination and protect against evaporation. We have used similar apparatus in studying the germination of conidia but for the study of mature penicilli and specific types of morphology, we discarded it many years ago on account of the atypical character of the structures produced when the colony was followed beyond the germination stage. The weakness of the method is clearly indicated in Ullscheck's descriptive notes where metulae and sterigmata are commonly "je zwei" "je drei," etc. Such numbers are rarely if ever characteristic of species in this group.

#### SELECTING THE INOCULUM

In an ideal culture system every colony would be grown from a single spore. In cultivating species of *Penicillium* attainment of this ideal is practically impossible. Various workers have used apparatus developed for the isolation of the single bacterium in order to secure their initial stock from one spore. No worker has recorded taking the same precaution for every subsequent transfer. Aside, therefore, from using a single spore culture method for obtaining the first colony, all have depended upon mass transfers for their subsequent studies. A transfer needle or loop thrust into a conidial area, then transferred to liquid or solid substrata carries many spores, hence most of the colonies resulting even from dilution cultures are composite growths from more than one spore. In spite of theoretical objections which may be advanced, we have species which have maintained their characters for twenty years under this system of culture.

As long as the stock available represents a single strain of *Penicillium* free from contamination by any other species of mold, yeast, bacterium or actinomycete, successive transfers in mass have been generally satisfactory. Such cultures can be easily maintained if handled in a culture room, which should be preferably steamed occasionally to carry down contaminations in the air, as well as protected carefully from gross currents of air. Where large series of species must be handled successively or miscellaneous moldy products must be examined, the probabilities of contamination are proportionately increased. In spite of care in transfer, an occasional culture will show colonies of another mold or abnormalities suggestive of impurity. Multiple transfers are a necessary precaution against the occasional contamination of this kind, but at best it is occasionally necessary to purify a culture of *Penicillium*.

The dilution method and single spore transfer have already been



discussed. Both are laborious, time consuming and involve many delays in the evaluation of results. For many years, however, our best results have been consistently obtained by multiple transfers from petri dish cultures. These have been most satisfactorily made with a short nichrome wire set in a smallbore tubular brass handle then ground upon a stone until the point is as smooth and sharp as a fine needle. This needle may be sterilized in the flame many times without injury beyond the occasional need of regrinding. With such a tool and a good dissecting microscope, or its 10  $\times$  lens held in the fingers, it is possible to select a satisfactory place in a conidial area, frequently a single penicillus and transfer a small cluster, even a few chains of conidia to a fresh petri dish. Again the very tip of a section of the mycelium free from all evidence of contamination may be cut off and used. This method of work has made possible the detection and identification of contaminations of many kinds and is constantly used to maintain our stock strains whenever questions of purity arise.

#### TEMPERATURE OF INCUBATION

Cosmopolitan saprophytes such as many of the *Penicillia* will grow well over a wide range of temperature. Thom (1910) reported that a range of from 15° to 25°C. produced only differences in the rate of development, not in the character of colony. Zaleski incubated his species at 17 to 19°C., with extreme limits 16° to 20°C., but reported no studies at higher temperatures. He reported correspondence with Biourge as suggesting 18 to 22°C. McCulloch and Thom found *P. gladioli* to present a very different colony at 14 to 15°C., to the one grown at 20 to 22°C., or higher. In our own studies of Zaleski's and Biourge's cultures, summer temperatures of 30°C., or higher in Washington, were found to reduce the growth of certain species to negligible while other species were not affected. The same general result has been several times reported by others. In general, incubation at 37°C., has been found unfavorable (Thom 1910) for general culture work but to be useful in separating certain species. Incubation between 18 and 25°C. representing the usual temperatures of laboratory working spaces is favorable for general culture of the group.

These experiences clearly indicate that the temperature of incubation should be part of the record in a complete and satisfactory cultural description. Temperatures have not been given in the past in describing *Penicillia* and are only given here for certain species but a temperature range of about 18° to 25° with a maximum under 30°C., may be assumed for all descriptions given unless other temperatures are specified.

## HYGIENE

Blochwitz, 1928, has correctly emphasized a whole series of conditions which must be considered in the study of molds. Temperature, humidity of the air in the culture vessel, water content of the substratum, favorable composition of the substratum and freedom from extraneous organisms. He designates organisms which grow well in liquid media as *hygrophiles* and those which fail to grow well in liquid media but grow well on dry areas, as *hygrophobes*. Whole series of vesicular cells, wound and twisted masses, or abnormalities may be traced to unfavorable physiological conditions or to the presence of specific organisms which act unfavorably hence may be called disease.

## PRECAUTIONS

Handling cultures of *Penicillia* inevitably distributes spores through the air of the room, over the hands, clothing of the worker, and upon every exposed surface. A rough calculation of the possibilities of certain well known species in petri dish culture gives a layer of conidia  $500\mu$  deep over an area 5 to 8 cm. in diameter. If those conidia are roughly  $4\mu$  in diameter, there are something like 75,000,000 to the square inch. Smaller numbers are more usual but those forms are not uncommon.

Exposure and rough handling of such cultures adds greatly to the difficulties of pure culture work. Discarded cultures should be destroyed by heat without opening. Petri dishes may go unopened into the steamer or into boiling water. Test tubes still plugged with cotton should go into the autoclave or the steamer before they go to the wash room.

If an inoculating room or culture chamber is to be built, it is desirable to finish the walls and ceiling with metal, tile or resistant enamel so that the room may be steamed from time to time, or if steam is not available, well sprayed with water or, better, with an antiseptic.

## GROSS IMPURITIES IN CULTURE

Major infections in morphologically widely different species are usually readily recognized and eliminated. But mixtures of related species, and minor contaminations with bacteria, yeasts, actinomycetes and sometimes hymenomycete mycelia introduce much greater difficulties because their presence is not so generally recognized by the worker who is handling the cultures.

## BACTERIA, YEASTS AND ACTINOMYCETES

Very close relations frequently exist between molds and bacteria, and a mixture may be transferred unconsciously many times. Yeasts present a closely similar problem, but greater difficulties are encountered with actinomycetes which sometimes seem to permeate the whole colony. The real dangers in such contaminations are due to the intimate relations which frequently exist between the two mycelia in the colony and which result in a composite but homogeneous appearance which may be carried through many culture generations. The exact relations existing have never been worked out, but presumptive evidence of parasitic habit has been frequently encountered in relations which sometimes seem to involve not only the vegetative mycelium but reach the single spore. Intensive investigation of these relations is urgently necessary as a part of our fundamental information as to symbiosis and parasitism.

The peculiarly penetrating odors of certain cultures of *Penicillium* isolated by ourselves and received from correspondents have frequently suggested the presence of Actinomycetes. Efforts to (see Westling) separate such organisms have usually failed as in the case of *P. bifforme* for example. Other cultures at first producing such odors have eventually lost that odor entirely although no Actinomyces was actually shown to be present. No. 4914a received from Gilman at Ames, Iowa, as *P. acidoferum* Sopp produced a very strong odor. Careful selection with the hand-lens disclosed a few mycelial areas in which the hyphae were well covered with mycelium with the appearance and measurements of Actinomyces.

Biourge reports the isolation of one of his *Oospora* species from Thom's no. 11, originally received from Thaxter, and carried for years as *P. luteum* Zukal. Similarly, we have found various cultures received from Biourge to be so far contaminated as to force us to question the validity of species based upon them. Westling's culture of *P. baculatum* as received from Stockholm contained a minor and submerged contamination of a member of the *Aspergillus repens* series which developed characteristically as soon as the species was planted upon agar containing a high percentage of cane sugar.

## PARASITIZATION (COMMENSALISM)

Certain species are repeatedly encountered in and among the hyphae and conidiophores of other species of *Penicillium* and even other genera. Sopp notes that the species of *Acaulium* (*P. brevicaulis*) are commonly

found growing and fruiting upon the mycelium of old cultures of other molds. We have made the same observation and noted the frequent presence of members of that group as entire replacements of other species in cultures received from other laboratories. Another group, the biverticillate *Penicillia*, presents a series of species or strains which are constant invaders of the colonies of other species. *P. rugulosum*, a member of this series, will overgrow and destroy cultures of *Aspergillus niger*, *Aspergillus tamaris*, and *Aspergillus flavus*. Members of this series have been found in the colonies of other green species. A pleomorphic culture of apple rot (*P. expansum*) received from California yielded *P. duclauxi* when carefully studied; another from Oregon yielded a form nearer to *P. luteum* and without the coremia.

A series of *Penicillia* from another laboratory showed two species of this same group in pure condition and two combinations that proved to be mixtures of the first two species. These experiences emphasize the necessity of close scrutiny wherever pleomorphic colonies are observed. Intimate mixtures, symbiosis and parasitism are very difficult to differentiate, but a rigid demand that every culture described be pure can not be overemphasized.

A culture received as *P. schneegii* and satisfying the description of Boas, produced elaborate sterile coremia which when transferred carefully proved to be an entirely separate species which appears to have been overgrown and largely submerged by the *Penicillium*. Separation of the two species made necessary a redescription of *P. schneegii* in a different section of the group (no. 330).

#### POISONING COTTON PLUGS

Entire elimination of mites by sanitary measures is possible but not usually attained. As a precaution in the preservation of stock cultures some one of the schemes of poisoning may well be used. One of these formulas consists of dipping the tips of the cotton plugs in a solution of 95 per cent alcohol—95 parts, bichloride of mercury  $\frac{1}{2}$  part, and glycerine 5 parts, colored with any of the aniline dyes. Care must be taken that the solution does not come in contact with the colony. The cultures must be allowed to develop into typical colonies before poisoning. An antiseptic formula for the purpose needs alcohol to insure penetration of the plug, a poison to destroy the mites, glycerine to prevent the crystallization of the poison as the alcohol evaporates, and the dye to insure the destruction of the cotton plugs when removed from the tubes.

## MITES

In a laboratory engaged in the study of the flora of raw products, mites are constantly brought in contact with the tables, the tools, the clothing and hands of the worker. Rigid isolation of the incubators, stocks of media, and lots of raw material brought in, with constant cleaning of the working space, tools, and the hands of the worker are the necessary price of preservation of purity in the cultures handled.

Mites travel slowly, but they keep going. They will migrate from petri dish to petri dish through a whole incubator and leave eggs, bacteria, and miscellaneous spores picked up en route wherever they go. If given time they go through cotton plugs, occasionally through paraffined cotton plugs, provided the culture within attracts them, for they seem to avoid some species and destroy all of other series. Mites in size are commonly so small as to escape notice by the naked eye. Workers with considerable culture experience have been found, who did not recognize mites, their odor, or their depredations until they were specifically pointed out.

## ANTS

Ants are equally capable of carrying contaminating organisms of all kinds. If the activity of either mites or ants was limited to the amount eaten, the damage would be less, but the culture is usually hopelessly contaminated after their visitation.

## DEGENERATION OF PENICILLIA IN CULTURE

Our collection of *Penicillia*, begun in 1904, still contains species isolated during that year (*P. camemberti* No. 5, *P. roqueforti* no. 18, *P. citrinum* no. 15, for examples). Other forms go back from ten to twenty years in continuous culture. Strictly conidial strains such as no. 5 (*P. camemberti* Thom) or no. 18 (*P. roqueforti* Thom) when transferred with rigorous care have come down through the twenty-five years unchanged. Cultures passed over to less critical hands for maintenance have often been lost, contaminated, or returned to us with marked changes of appearance believed by us to represent the "introduction" of parasites, commensals or contaminating species although it has not always been possible to prove the charge. Loss of ascospore production by no. 4010.7 (type strain of *P. awellaneum*) was presumably due to the presence of both of a pair of conjugating haplonts in the original culture and the gradual but unintentional elimination of one of them by selective transfers. In several biverticillate forms there have been changes in cultural habit;

for example, no. 2670 (type strain of *P. purpurogenum* var. *rubri-sclerotium* Thom) seems to have lost its sclerotium production several years ago, so that that form, as utilized by Herrick and May in the production of gluconic acid, is scarcely recognizable as the same species.

#### LONGEVITY IN CULTURE

The preservation of cultures over long periods without loss of type species has been much discussed. Strains of *Penicillium* differ greatly in their ability to withstand dessication. In keeping a large collection in the ice-refrigerator at about 10°C. (50°F), transfers upon wort or Czapek "slants" have been found necessary once a year to avoid the occasional loss of the more delicate species. Many species survive for several years. Extensive series of transfers have been made to reach a basis for generalization but there are too many factors involved to generalize safely. In handling *Penicillia*, our practice seems to justify keeping three series, one at laboratory temperature, one in the ice-refrigerator and one in cold storage at or near the freezing point. Aside from an occasional delicate species the "cold storage" stock has proved the most dependable.

When cultures fail to respond on the first transfer, the possibility that some viable spores remain is often realized by Povah's method which consists in flooding the surface of the culture with melted agar.

#### STALING EFFECTS

If the medium in a petri dish is thickly seeded with the spores of one or several *Penicillia*, the entire surface will become densely covered with mold, showing here and there, perhaps, the dominant features of particular species. If only a few colonies of one species are widely spaced in such an area, the resulting colonies may either blend or cease to develop. When such colonies blend there is no line of demarcation. When, however, they cease to develop as they come within the zones of influence of adjacent colonies, lines are maintained free from mold between these colonies. A normal spread of mold growth over the substratum takes place in both colonies outside of this free line or zone. Similar relations are found between colonies of different species and of different genera. They may grow together or inhibit each other and will repeat the same picture in successive cultures.

Another phase of the same general phenomenon is seen in colonies which cease growing while wide areas of the substratum are yet free from mold growth. In such instances the spores of the species may be

scattered over the free area and yet fail to germinate. In contrast, other species presenting the same proportions of colony will show the surrounding free area thickly set with young colonies.

If young mycelia, coming into immediate contact with each other combine to fill an entire free surface without showing colony lines, the inference may be drawn that stages in life history producing inhibitory substances have not been reached. Such lines of demarcation evidently only occur when colonies approaching each other are old enough for "staling" to have occurred.

Great contrasts occur between the effects of different species as they come within these zones of influence. Some combinations result in increased growth, some produce bright colored areas in the reverse of mycelium or substratum, or both. No satisfactory explanation has been given which will apply widely to these effects, although several workers report extensive experimentation.

Harder based his conclusions merely upon observations on the rate of growth and color production in the medium and mycelium. He attempted to determine whether inhibition or stimulation of growth might not be the result of depletion of available carbohydrates in the medium or a change in hydrogen ion concentration. The investigations of Zellar and Schmitz with wood-destroying fungi and *Aspergilli* show that there were not as many instances where one colony grew over another as there were of inhibition of growth of one fungus before or after contact with another. These results are in agreement with the general observation that fungi in their growth show a more marked tendency to grow out and away from the medium influenced by their own growth metabolism than to grow towards a center containing diffused materials.

Pratt, in 1924, surveyed the literature on staling of fungi in culture. Using the *Botrytis* germ-tube method, she studied Richards' solution on which *Fusaria* had been grown, and states that alcohols and acetaldehyde are toxic only in high concentrations, while simple organic acids are toxic in sufficiently low concentrations to be responsible for poor germination in an acid, but not an alkaline medium. Staling is concluded to be due to the formation of bicarbonate by the carbon dioxide of respiration whenever the medium is of such a nature that a basic radical is set free. Species of *Penicillium* used in germination experiments responded similarly to other fungi.

## CHAPTER VI

### OBSERVATION AND DESCRIPTION OF *PENICILLIA*

The student of a mold has before him a colony with certain striking characters, such as color, and color changes, floccosity, conspicuous structures, such as coremia, perithecia or sclerotia, contour or spreading habit, its relation of discoloring or liquefying the substratum, the production of conspicuous drops of transpired fluid, odor, etc. After a preliminary survey with the naked eye, the hand-lens and direct examination with the low magnification of his compound microscope, a microscopic mount enables him to determine the details of structure and measurement of essential cells.

#### SLIDE MOUNTS

Preparations for microscopic examination present many difficulties. The aerial parts of many *Penicillia* do not mix readily with water. A portion of a colony transferred to an aqueous solution may float for days without penetration by the fluid. Occluded air accounts for only part of this effect. In experiments with fixation for cytological work, vials of such solutions were subjected to 26 inches of vacuum for more than twenty-four hours without penetration by the liquid. Wehmer demonstrated (personally) his method of work by taking a portion of a colony with the forceps and washing it in a watch glass of alcohol, then teasing portions of the mass for examination. After trying many methods our best results have been obtained by removing from the colony small masses perhaps 1 to 2 mm. in diameter, selected under a hand-lens as favorable for study, placing them on the slide, allowing a drop of alcohol to flow across the mass followed quickly by water, or mounting fluid. For mounting, water is satisfactory if observations are to be completed in a few minutes. Weak glycerine (10 to 20 per cent) with or without such a stain as eosin can be used over a longer period or allowed to concentrate, then sealed for a semi-permanent preparation. A mixture of equal parts of 2 per cent aqueous potassium acetate and 40 per cent glycerine colored with copper acetate makes satisfactory mounts for examination. Shrinking or swelling in the preparations used for examination probably accounts for some of the discrepancies between measurements reported.



All of the observations reported by us have come from fresh mounts made from cultures under observation in the laboratory. Preparations from the edge of the growing colony have been regularly supplemented by mounts from older parts of the colony in which ripeness of the spores has been assured.

Macroscopic observations on *Penicillium* should begin by the third day and follow the development of colonies regularly through the growing period usually comprising the first two weeks, followed by occasional records of the changes in color, appearance of the colony and its effect upon the substratum over a period of several weeks. Microscopic studies should include the submerged and aerial hyphae, the conidiophores, and the entire conidial apparatus together with the natural arrangement of its elements in the penicillus.

#### EQUIPMENT

In making such observations a good hand-lens is useful for the macroscopic survey; for the closer examination "plate" or petri dish cultures capable of direct observation under the lower magnifications of the compound microscope are essential to an accurate description of the colony itself. A good objective and proper oculars, with magnifying power up to about 1000 diameters, together with a standardized micrometer, are essential in detail studies of the ultimate cells, in determining the methods of spore formation, the markings of cell walls and the connections between the various cells involved. For this purpose, we have used Zeiss Apochromatic objective 16 mm., 8 mm., and oil immersion 3 mm. apert. 1.30, associated with a 12× Compens ocular, in which an eyepiece micrometer disk makes constant provision for measurements to be made directly.

The specific observations desirable in describing a species of *Penicillium* must be discussed in detail in the order in which they are available.

#### COLOR OF THE CONIDIAL AREAS

The first character to strike the observer of one of these species is the colony color. Every description of a *Penicillium* emphasizes some color or series of colors as characteristic. If the describer has used several culture media, he commonly finds it necessary to specify the color upon each by a name or reference to a different number in some series of color standards. Our own records over many years show that the same strain will be described by several names from Ridgway in as many years. In general, however, the colors produced by any strain of *Peni-*

cillium upon a standardized substratum fall fairly consistently within a narrow range of tints and shades of some particular mixture of yellow and green, blue and green, red and orange, orange and yellow, etc. The culture medium used must, however, be the first item specified if the color ranges which follow it are to have real value.

The range of colony colors within the group includes a few white forms; various mixtures of orange and yellow, with admixtures of red in one series; and a great series of forms, yellow green, green and blue green during their fruiting period, many of which lose all their green or blue color in age, thus becoming various shades of yellowish brown, reddish brown, purplish brown, to almost fuscous in age. In none of the typical *Penicillia*, however, are these colors accompanied by the heavy brown walls of the *Dematiaceae*.

In a general way, green and blue green colors are linked with fermentative activities in which nutrients are decomposed with the production of acid by-products. Similarly the brown colors of old colonies are more or less definitely linked with products of alkaline nature. Demonstrations of exact relations of acidity and alkalinity have not been successful. Cultures in the presence of such indicators as litmus, phenolphthalein, and some of the hydrogen ion indicators of Clark and Lubs have shown the general trend of color changes to follow the fermentative activity of each mold.

#### COLORS OF THE MYCELIUM

The aerial mycelium in *Penicillium* is nearly always colorless. Yellow and red appearing hyphae and areas are common in the biverticillate series, but when examined with the microscope the coloring substance is found deposited as granules on the surface of the cell wall, not within the cells.

On the other hand, the submerged mycelia of various species and groups show a brilliant series of yellow, orange, red, lilac or hyacinth colors and combinations of colors, with occasional green, brown, and even black areas. These colors are seen from below or by turning over the petri dish or tube and have been designated as "color of reverse" or "reverse." Culture of many series of species on a wide range of nutrients shows that the color production of a species may be markedly influenced by changing the nutrient often without apparent injury to the vigor of the mold. Color may often be increased greatly by increasing the concentration of some fermentable nutrient, or again entirely inhibited by the absence of that particular substance. In other words,

many of these colors are by-products of a metabolic activity of the mold which may be transient and evanescent or which may accumulate in the hyphae until they themselves are densely filled with the coloring substance.

A succession of colors in the submerged mycelium is characteristic in many of these species. The mechanism of color production is obscure and unsatisfactorily known, but in cultures of *P. duclauxi* upon sucrose media the initial colors in the mycelium are rich yellow to orange and the reaction is definitely acid; in the older colonies the reverse becomes red and the reaction is alkaline. A drop of alkali inserted in a yellow area produces an immediate change to red; a drop of acid in contact with a red area brings back the yellow color. While very few of the coloring substances found are in this way reversible, the general fact that yellow colors in the mycelium are associated with acid conditions and red colors with alkaline conditions has been established fairly well.

The part played by oxidation in these successive color changes has not been worked out systematically but is indicated as considerable by isolated observations. In cutting Roquefort cheese containing a pure culture of *P. roqueforti* the mold is sometimes seen as yellow but to change to green within a very few minutes. In a culture of the *P. luteum* series in which the reverse was entirely yellow, a piece was removed for microscopic study and the area surrounding the hole in the agar became quickly red. Progressive changes from red to yellow shades are constantly seen in slanted tubes and in petri dishes with uneven thickness of substratum. The mechanism of these changes is totally unknown but appears to be partly at least dependent upon contact of oxygen with the pigmented material.

#### COLOR IN THE SUBSTRATUM

The coloring matter already discussed as present in the mycelia may or may not escape into the substratum. In some species, the substratum is quickly discolored and follows the color changes of the mycelium. In others the color is retained in the cells of the hyphae. These contrasting conditions may occur in nearly related strains, leading to the hypothesis that the difference is a minor change in biochemical condition rather than having fundamental importance.

These color differences are so conspicuous in their contrasts that they have been repeatedly emphasized in classification without always recognizing that the course of color change in a series of species or strains presents a gradation from one extreme to the other in which one par-

ticular strain beginning at the common starting point seems to carry a color production process to a particular point represented by a determinable tint or shade, and stop. Another related species beginning at the same point may carry the same process a trifle further and a third still further. Thus in the *P. luteum* group we have forms with the mycelium for a long time yellow, others which pass quickly through yellow to orange and stop, and still others which become first yellow, then orange, then red, and finally some in which the yellow and orange are transitory and the red predominates so early in colony development that the yellow shades are overlooked except when great care is given to the observations.

Such considerations as these, brought about the recognition of series of related forms in which great color contrasts occur, but which were found to present consistent morphological characters. Thus Westling described his *P. viridicatum*, giving the specific color ranges in conidial areas and in reverse. With his strain (our no. 2552) in culture, we quickly found a whole series of forms with fairly consistent morphology shading into each other in color reactions but sharply contrasting when individuals widely separate in the series were compared. Among Biourge's species certain of these organisms far enough apart to show color contrasts are found described as different species without their indisputable relationship as a series being clearly brought out.

Investigations of the pigments produced by various species of *Penicillium* have covered the solubility and routine reactions of the coloring matter of *P. herquei* (by Sartory and Bainier, 1911), of *P. divergens* and *P. citricolum* and five species of *Citromyces* (*C. minutus*, *C. ramosus*, *C. cesiae*, *C. musae*, *C. cyaneus*) in 1913 by Bainier and Sartory. In their papers the pigments discussed showed more or less differences in solubilities and reactions characteristic for species and groups of species. The chemical nature of the pigment was not worked out for any species. Enough was done to show that a field of theoretical chemical interest might be developed but little work has been done to determine whether any practical applications might be developed. An unnamed species was studied chemically by Martini and Déribéré-Desgardes in 1914.

#### APPEARANCE AND TEXTURE OF COLONIES

The appearance and texture of the colony are among the first characters to be seen. Examination with the naked eye, the hand-lens and low magnifications of the compound microscope establish four extreme types of aerial or surface growth, velvety (velutinous), floccose, coremi-form (or fasciculate) and ropy or funiculose.

*Velvety (fig. 7)*

Colonies are typically velvety or velutinous if all or nearly all of the vegetative hyphae are submerged in the nutrient substratum (example, *P. digitatum* Sacc. upon Czapek's solution agar) and conidiophores only, branching from submerged hyphae, rise above the surface and produce



FIG. 7. Diagrammatic radial section of a *velvety* colony; magnified 25 times: *ab*, surface line of substratum; *c*, conidial area; *cp*, conidiophores standing approximately parallel and running from submerged mycelium, *sm*, to conidial layer; *f*, margin or fimbria.

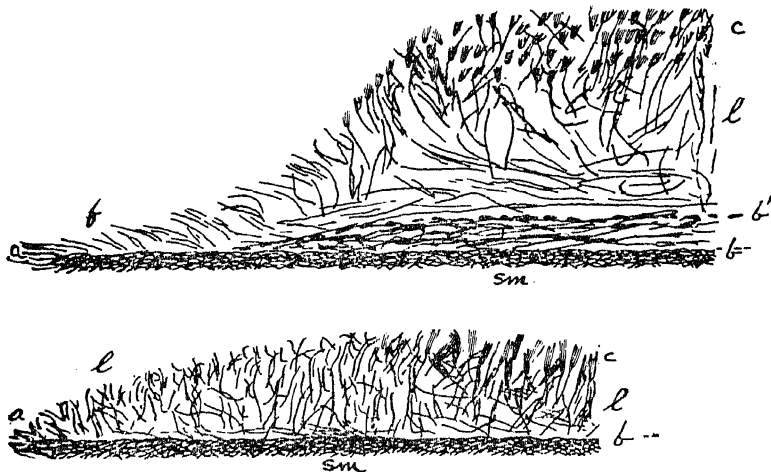


FIG. 8. Diagrammatic radial sections of two *floccose* or *lanose* colonies; magnified 25 times: *ab*, surface line of substratum; *c*, conidial area; *l*, floccose hyphae; *sm*, submerged mycelium.

the conidial masses. Such colonies give the appearance of a surface of velvet or a field of grain such as wheat.

*Floccose (fig. 8)*

Other species, for example, *P. camemberti*, produce a white cottony mass of branching and interlacing hyphae spreading evenly or unevenly



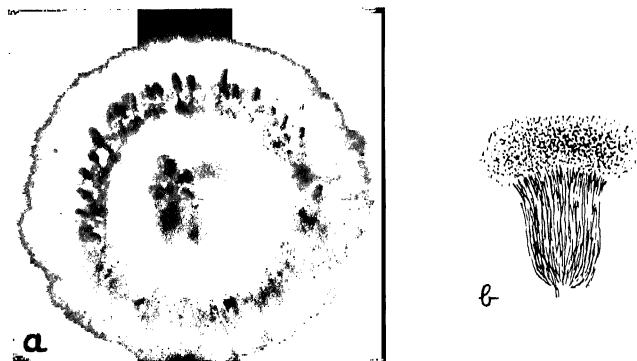


FIG. 9. Coremium formation (a) from photograph of *P. clariforme*; (b) single coremium of *P. expansum* in special media.

over the surface of the nutrient medium and indistinguishable from vegetative hyphae. At a characteristic period in the development of such a species, conidiophores appear as branches of these aerial hyphae. Colored conidium-producing areas usually appear first in the center of the colony and gradually spread toward the margin. Throughout the growing period a sterile white or in some species a yellow or reddish margin surrounds the fruiting area. When growth ceases the fruiting area commonly extends to the very edge of the colony.

In their extreme forms velvety and floccose colonies are sharply differentiated; in actual study of cultures every gradation between these extremes is found. In *P. roqueforti* and its allies the mycelium spreads broadly at the very surface of the substratum, alternate sections of many of the hyphae developing above and below the surface. The next step in complexity involves a branching and interlacing network of mycelium closely applied to the surface of the culture medium and the production of the conidiophores mostly from this basal network; such colonies, however, have the same velvety appearance as the typical velvety form. For descriptive purposes an arbitrary application of the terms has been used in which colonies showing a definite aerial felt or mat before conidium formation becomes evident have been called floccose; colonies in which conidial areas follow closely the growth of the mycelium at the margin and give in general a velvety appearance have been called velvety; modifying terms have been introduced where necessary.

#### *Coremiform (fig. 9)*

In a few species separate conidiophores, if present, are only found by microscopic examination. The colonies consist of a submerged vegetative mycelium and upright coremia (columns of hyphae or conidiophores) giving the macroscopic effect of a stalk and conidium producing head. Those extreme types in which the simple penicillate types of conidiophore did not appear in the specimens studied, have been described in the literature from other characters under the names *Coremium* Link, *Stemonitis*, *Stysanus*, *Isaria*, etc.

#### *Coremium formation*

Various authors have discussed the phenomenon of coremium formation in *Penicillium* and related genera (Hallier, Brefeld, Sopp, Wehmer, Thom, Wächter, Westling, Boas, Munk, Schaposchnikow and Manteifel Blochwitz, Trzebinski, Schilberszky). As reported, coremia are constantly produced by certain species, sporadically produced under unde-



finer conditions by others, produced in particular substrata or concentrations of substrata by other species.

Some hold that coremium formation may be induced in any *Penicillium* by proper culture methods; others that coremium formation is an inherent characteristic of certain species and becomes evident only if conditions are favorable. Our own observations covering many thousands of preparations indicate that coremia may arise in connection with either of two forms of antecedent structure, (1) the development in the mycelium of trailing or ascending ropes of hyphae which frequently anastomose in characteristic manner, and, (2) when mycelium without other divergence in general appearance produces all or part of its conidiophores in clusters or fascicles (tufts) with or without sterile or partially sterile areas between. The accentuation of either tendency leads directly to the structures described as coremia, but the structural background in cultures of these and frequently of related species in which typical coremia have not been found, persists in whole series of forms and is fairly readily recognizable. Thus the interlacing ropes of hyphae found in the *P. brevicaulis* (*Scopulariopsis*) group and a section of the *P. luteum-purpureogenum* series resemble the structure seen in cultures of *Isaria* (see Atkinson) by ourselves and others, while the other type of structure typified in *P. expansum* is that which characterizes the genus *Coremium* of Link and of Corda's *Icones*. We have not seen satisfactory evidence of coremium formation induced in species which lack one or the other of these antecedent mycelial structures, but species showing these structures do regularly or sporadically produce coremia.

*The fasciculate series (fig. 10)*

When transferred to laboratory culture media a few species retain the strictly coremiform habit (*P. claviforme* Bainier or *Coremium silvaticum* Wehmer, for example). Other species produce conspicuous coremia in particular situations. *P. expansum* commonly produces coremia upon stored apples without obvious development of simple conidiophores, but it produces large numbers of ordinary penicillate fruiting hyphae in the dilute media of the laboratory with more or less conspicuous coremia varying with the strain studied. *P. duclauxii* Delacr. on bean agar is a simple *Penicillium*; if sugar is piled upon one side of the colony, coremia quickly appear and dominate the subsequent growth occurring. In a whole series of species of which the apple rot *Penicillium* (*P. expansum*, *P. leucopus*, *P. glaucum* of various authors) is the best known, part or all of the conidiophores arise either from submerged hyphae or from aerial

networks in upright groups, clusters, or fascicles, which occasionally become typical coremia. The presence of these fascicles of conidiophores in a conidial area otherwise usually velvety in appearance gives an irregular or at least an uneven appearance to the developing margin of the colony. This has been designated rough or mealy in Westling's descriptions. These fascicles commonly form taller clusters and produce larger masses of conidia than the simple conidiophores which in many strains and in our ordinary media, more or less completely fill the intervening spaces.

*"Rough or mealy" margins*

A whole series of forms show this *rough* or *granular* or mealy marginal area in the rapidly growing colony. Microscopic examination in such cases confirms the interpretation that fasciculation of conidiophores is the cause of the rough or mealy appearance. Studies of this group continued for many years and under many cultural conditions indicate that this whole series of species share to some degree the ability to produce



FIG. 10. Diagrammatic radial section of the margin of a fasciculate colony; part of the conidiophores are produced in clusters or fascicles giving a "rough" or "mealy" appearance to the surface.

definite coremia when proper conditions are found. Conversely the production of coremia of the *P. expansum* type, that is, as aggregations of conidiophores into a stalk from which the tips diverge at the apex to produce the usual penicilli of the group, is limited to the species in which the initial fascicles are regularly present and evident to naked eye or hand-lens examination at some stage of colony development. In some of these species the fascicles noticeable in the marginal area are later so crowded by the development of the simple conidiophores as to be no longer recognizable. In a few cases, coremiform fascicles are noticeable only at the extreme margin of the colony and comparatively late in colony development as in *P. italicum* Wehmer. In other species, the vast majority of the conidiophores are bound together more or less closely into such fascicles as are seen in *P. granulatum* Bainier, or some strains of *P. expansum*. Every gradation between such extremes can be found when hundreds of *Penicillia* are brought together and studied.

*Ropy or funiculose surfaces (fig. 11)*

Some species are characterized by aerial ropes or bundles of several to many hyphae, branching and interlacing over the surface, ascending, but rarely, if ever, upright (vertical). In these species part or all of the fertile hyphae arise as branches from these ropy networks, although usually simple conidiophores are also found. This type of structure occurs in at least five subgroups of this great series of forms, the monoverticillate, the biverticillate, polyverticillate, *Scopulariopsis*, and *Paecilomyces*.

Ropy masses of aerial hyphae are readily determinable with the lower magnifications of the microscope and form a useful character in grouping these species. They do not seem to link together the large groups in which they are found, but within these large groups such ropes are a useful diagnostic character. Guesses as to genetic significance would be unprofitable.

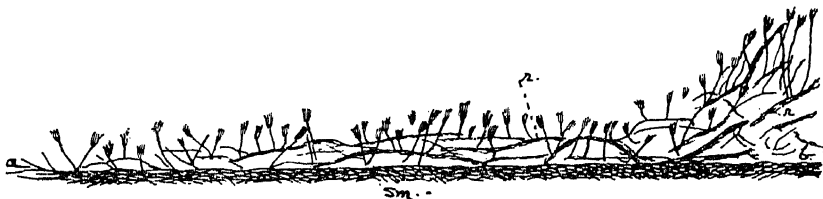


FIG. 11. Diagrammatic radial section of a funiculose or ropy colony (*P. pinophilum*); magnified 25 times: *ab*, surface line; *r.*, ropes of aerial hyphae.

*The margin (Fimbria in Zaleski)*

For most species correct information as to the relation between submerged mycelium and conidiophore production can be found only at margin of the growing colony. The period of satisfactory observation thus begins about the third day for rapidly growing species and ends, as a rule, with the cessation of rapid growth within the second week. In some species the mass of mycelium is so great even at the margin as to interfere with satisfactory use of the compound microscope. Often, however, by selecting petri dishes in which two or more colonies are present the necessary observations can be made in the area of inhibition between two colonies in which a growing zone normally wide is greatly narrowed.

The extremes of habit observed prominently in the study of marginal areas are represented by broadly spreading colonies which quickly cover all of the surface of the substratum and by the narrowly growing

species in which a narrow border of submerged mycelium is quickly followed by conidial areas, the colony rarely, if ever, becoming very large. In making such observations the extremes are conspicuous and readily characterized; for the intermediate forms necessarily other characters are much more useful.

### *Zonation*

Surface growth in zones is a conspicuous feature in many species. It was discussed extensively by Munk in 1912. Biourge in his subgenus *Eupenicillium* makes zonation the diagnostic character of Subsection I, and puts in his hemizonate series of that subsection forms indistinctly or indefinitely zonate. It has seemed to us in the examination of Biourge's descriptions and his cultures that this basis of grouping has thrown together too many forms of divergent relationship. Zonation valuable because conspicuous and fairly constant for certain species is an unanalyzed growth habit occurring in different sections of the whole genus and may better be linked with other characters than used as a single basis for establishing a taxonomic section. In some species zone production is only transiently evident at early stages of colony growth; again it shows only at the latter end of growth and in inconspicuous degrees. Some species are zonate when grown upon one substratum and not on other substrata. It has seemed best, therefore, to disregard Biourge's zonate section and use zonation in the separation of members in series held together by what we regard as more fundamental characters.

Ullscheck following somewhat the arguments of Munk, offers the hypothesis that zonation appears when colonies grow rapidly, secrete enzymes and produce by-products of their metabolism of such concentration in bands of the nutrient as to reduce or suppress growth and fruit formation in those bands. The mycelium advancing into fresh nutrients resumes vigor of growth and abundance of fruit formation only to be depressed again by the excessive by-products of this heightened activity. The extension of the colony thus gives the appearance of zonation.

### *Drops*

Transpiration of fluid is a conspicuous feature of some growing colonies, evident but inconspicuous in others and not evident in still others. Where transpired fluid accumulates in drops, it becomes a conspicuous feature of the growing colony with characteristic color. Evaporation in the older colonies frequently leaves residues upon the surface of the conidial area and depressions in the surface where conidial

masses were pressed down by the weight of transpired droplets. These residues and depressions must be correctly interpreted. One disturbing factor introduced by these drops is the germination of conidia in the transpired fluid. This occurs in some species and results in globose masses upon the surface of the older colony which may or may not produce secondary conidial areas. While the specific studies recommended, emphasize the structures at the margin of the colony, these mold balls due to secondary growth have been occasionally misinterpreted.

### *Odor*

Characteristic pungent or penetrating "moldy" odors are produced by many species. Nomenclature of such odors is so utterly unsatisfactory that we have been entirely unable to interpret most of the terms used. Penicillia are responsible for much of the odor designated "moldy," they share with the Actinomycetes responsibility for the term "musty." With the failures of our descriptive terms, about all that can be accomplished is to indicate the presence or absence of an odor and its intensity with an occasional descriptive simile which may help part of the users interpret their findings.

### CONIDIOPHORES

The essential data as to conidiophores are their length, septation, the diameter of their cells, and especially their origin and relation to the substratum and to each other. The walls of the conidiophore may be smooth and thin, or may be pitted or may bear concretions or warts. These differences in the cell wall run consistently through certain series of species, hence must be observed carefully with the best objective available. Although extremes of variation in length of conidiophore may be very marked in any culture, the majority of conidiophores in any such culture approximate an average length. This length to be most reliable must be taken from the origin in another hypha to the lowest branch of the penicillus. If the penicillus were counted into the length, the length of conidiophore would in many cases be doubled with the maturing of the spores. The actual length, however, is little changed with such maturity. Valid data on these points can be secured in many species only by direct observation of the undisturbed colony in the air under the microscope, instead of by the study of fluid mounts. This is equivalent to saying that petri dishes, or other vessels which can be uncovered for study, must be used. The student must expect, therefore, to make many cultures and jeopardize the purity of one such culture every time he undertakes its proper examination.

The conidiophore of *Penicillium* lacks the differentiated footcell so characteristic of *Aspergillus* as is indicated by Thom and Church in "The *Aspergilli*." While species with difficultly interpretable structures are found the distinction remains generally definite and rather easily demonstrable. Earlier authors failed to recognize this difference although it was figured by Ferdinandsen and Wing as a specific character for *Sterigmatocystis dipus* without recognizing its occurrence in the whole genus. As a result we find monoverticillate *Penicillia* sometimes included in *Aspergilli*, again (Biourge) *Aspergilli* included in *Penicillium*. Gilman and Abbott (p. 283) accepted the distinction as drawn by us yet contributed to us as *P. albidum* Sopp, our no. 4894.4, which in our hands proved to be a member of the *Aspergillus nidulans* group. The question whether Sopp may have included various species of the "Microaspergilli" among his sixty species of *Penicillium* many of which have never since been recognized, is valid but probably unanswerable.

#### THE PENICILLUS

Biourge uses the term *Penicillus* for the whole conidial apparatus (Pinsel in German), which he measures as beginning at the lowest branch upon the main axis and including the tips of the sterigmata. Conidial chains are excluded because they vary in length with the age of the colony, whereas the branching system does not. To have value for descriptive purposes the range of variation in the branching system rather than the measurement and description of some particular penicillus must be described. More important still the type of penicillus most common in a species can best be determined by study of preparations under low magnification which makes possible the rapid comparison of large numbers of penicilli. This can be done most readily by direct examination of the colony in the petri dish and habit sketches can be made with the camera lucida to show the relative size of penicilli, their arrangement, the types of branching observed, and the arrangement and course of the conidial chains (see fig. 2, Chapter I).

In describing species the general type of penicillus presented must be noted as monoverticillate if each terminal branchlet with its cluster of sterigmata and conidial chains seems to stand out separately and to conform to the same particular type; when recognized as monoverticillate, (a) it is strictly so (stricta), (b) borne upon one of an irregular series of branches at various levels upon a common fertile hypha (Chapter XIII) or (c) on one of several diverging branchlets from the tip of the main axis (Chapter XVII); (2) as biverticillate, if branching occurs at two

levels, and symmetrically biverticillate if the two groups of branches are regularly and evenly spaced about the center. If the penicilli appear to be asymmetrically biverticillate the species is sought in the divaricate section of the monoverticillate group, or among the asymmetrica (3) in which the branches are characteristically alternate or form incomplete whorls about a central axis. In any case the description should indicate the arrangement of parts and the number of series in the branching system as characterizing the species.

In published descriptions of penicilli many difficulties are encountered. No standardized description has been found. Biourge in dealing with branching systems reports branches in pairs when one is a branch and the other a continuation of the main axis. A careful study of his figures for species whose type strain we have in culture shows that his numbers of branches, metulae and sterigmata while manifestly based upon his drawings, hence upon the particular penicilli drawn are not characteristic nor fixed in the species. In fact, wherever checked against actual cultures the numbers have proved entirely misleading. Similarly, it is not possible for a "penicillist," to use Biourge's term, to believe that Oudemans had a member of this group with metulae and sterigmata in threes (see *P. humicola* Oud.) whatever he may have seen in the particular slide he studied.

There is nevertheless a general type, number, arrangement and size of elements in these species which can be described in general terms and figured to some degree in a habit sketch. In a detail drawing it is only possible to show a few characteristic cells in one or two penicilli.

In the representation of the penicillus, we find in the literature a wide range of practices; Corda idealized his picture; Brefeld after careful study of his preparations prepared a series of diagrammatic shaded figures giving his own conception of the production of branches, metulae, sterigmata and conidia. Wehmer seizing a bit of a colony with his forceps washed it free as possible from conidia in a watch glass of alcohol, then mounted it for examination upon a slide. Biourge used cultures preserved in alcohol, teased carefully to present the utmost detail of structures remaining. For the detailed study of the origin, shape, measurements and markings of the cells in the penicillus some such mount is necessary and the figures of Wehmer, Westling, Weideman, Biourge and Zaleski show valuable details but many of these line drawings show little regard for these as plants with living constituents composing their many related cells. The relations as pictured between metulae, tips of sterigmata and conidia break every law governing the peculiar internal

structure of fungi. To one who has not only made thousands of such preparations and studied them, but has studied thousands of cultures in petri dishes where every element of the colony could be seen as it stood undisturbed upon the substratum, much valuable information is lost when slide mounts alone are studied. The detail drawings are desirable, but the habit sketch made by direct observation is also essential to any proper placing of the organism among its fellows in the group.

#### METULAE

The metulae of a species follow closely the diameter of the main axis and its branches, though they are commonly a little smaller in diameter. When the walls of the main axis are pitted or rough the branches and metulae may or may not be rough. Their arrangement and length are usually fairly characteristic of the species. The variations in shape are such as may be easily attributed to the effect of crowding many such elongated cells into compact verticils upon the apex of the fertile branch.

#### STERIGMATA

The sterigma as the typical conidium producing cell of the whole group presents the most specialized cell of the whole plant. The first sterigma is borne on the tip of the main axis; a little to one side of its base the second buds out; similarly the other sterigmata of the verticil bud out consecutively and with more or less regularity from the same fertile area. There may be few to many in the verticil. Typically a sterigma is a cylindrical cell narrowed at the apex to a specific diameter to form the characteristic conidium-bearing tube of the species and cutting off conidia successively from its tip so that the resulting chain of conidia has fully ripe cells at its distal end and developing cells at its base sometimes until one to several hundreds are present in the chain. It is therefore to be regarded as a differentiated propagative cell differing from all other cells in the plant body by its capacity for repeatedly putting out new cells from a specialized spore-producing apex or tube.

#### *Shape of sterigmata*

Different forms of sterigmata characterize certain sections of this great group. The sterigma of most of the monoverticillate series and the *Asymmetrica* is a cylindrical cell (fig. 12) with an acute tapering apex to form a tube roughly half the diameter of the sterigma. In the biverticillate series the sterigma is proportionally smaller in diameter and longer, narrowing more slowly (acuminately) at the apex to a much smaller



tube (perhaps one-third) compared to the diameter of the main sterigma. In the soil *Penicillia* the tapering of the apex is commonly more abrupt and carried to a smaller tube so that descriptions of species in this section read "beak like" sterigmata. In the *Paecilomyces* (*Penicillium divaricatum* Thom) series of species the sterigma consists of a broad short basal tube narrowed to a long neck which is bent at its base from the main axis of the sterigma. In the *Scopulariopsis* group (*P. brevicaulis* and allies) the sterigmata are sometimes *Penicillium*-like but the tubular character is commonly lost in a form in which the cell begins at or near the base to taper gradually to the diameter of the newly forming conidium. In some strains the sterigma is narrow at the base and forms a straight tube producing conidia at its apex.

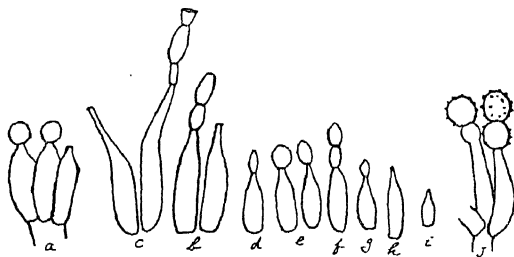


FIG. 12. Sterigmata: *a*, the acute type; *b*, the acuminate type; *ig*, *hi*, the "beaked" variety of acuminate sterigmata; *j*, the *Scopulariopsis* type; *c*, the *Paecilomyces* type with its long tube turned sharply away from the axis of the cell.

#### CONIDIA

Every conidium arises theoretically as a cylindrical body cut from the tube-like tip of a fertile cell (sterigma). Changes from the cylindrical form may begin before the separation of the cell has become evident by a definite wall, or may be delayed. In a few forms such as *P. italicum* Wehmer, the cylindrical shape persists for considerable time to be gradually transformed to elliptical. In the biverticillate group the conidium long-cylindrical at first and small in diameter tends to become fusiform by rapid increase in diameter at the center. In some of these species the globose condition is reached by continued swelling. In the Eupenicillate series (of Dierckx and Biourge) and the monoverticillate series the initial diameter of the cylindrical conidium is commonly much larger than in the biverticillate series, the segment cut off as the new conidium, varies in length in the different species. In some forms the segment is long,

therefore, a very definite ellipticity persists through the ripening of the conidium. In other species the length of the segment is little greater than the diameter, hence the conidium quickly becomes globose or sub-globose. Although conidia in certain species have been described as globose at first, becoming elliptical in maturity (Westling, p. 84) no evidence of this condition has ever been seen by us, hence we are compelled to believe that the report was based upon misinterpretation of observations. This may easily arise. In very rapidly growing forms the change from the initial cylindrical form passes so quickly that conidia in this condition are only found by very careful and often prolonged observation. If the same species is studied from old colonies in which the rate of development is much slower, the cylindrical and elliptical forms persist for much longer time or even replace the globose in some preparations.

The conidia in *Penicillium*, form unbranched chains which may contain few to many cells. A conidium usually attains the characteristic size, shape and markings for the species by the time a half dozen newer conidia have been cut off between it and the sterigma. Walls may thicken some, markings may accentuate a little, but otherwise little visible change occurs after that time even though the conidium may remain attached in the chain for a considerable time.

#### *Phenomenon of Corda*

Corda in describing and figuring his *P. fieberi* (see no. 574) specified that the end conidium of the chain, or the oldest and the second oldest conidium in the chain, were much larger than the others. No other observation of this kind was recorded until Biourge (monogr.) reported the "phenomenon of Corda" on page 308 for *P. cinerascens*, on page 299 for *P. aureo-flavum*, on page 284 for *P. chermesinum*, and on page 282 for *P. aurantio-violaceum*.

In a culture (4991) belonging to the *P. brevi-compactum* series, the picture given by Corda was very beautifully reproduced. In this culture drops of fluid were exuded at various points in the conidiophore and sometimes in the penicillus. These drops varied from very small to large and in certain cases developed about the conidia at the very tips of part at least of the chains. A sketch drawn from direct observation of such a penicillus would have given Corda's figure but probably does not give any real clue to Corda's species.

Close scrutiny of our own cultures of these species and of the whole group leaves us little ground for agreement with either Corda or Biourge.

In such chains of conidia as we find in *Penicillium*, the individual spore after it reaches its full size, shows only slight changes in form, while rounding up, and in the development or accentuation of markings. No significance can possibly rest in the ultimate or penultimate position in the chain unless these cells come in contact with the medium or with some other source of moisture and enter the swelling stage which precedes germination in most species.

In this respect, species differ greatly. Many, perhaps most species either by their "staling" effect upon the substratum or by drying the area invaded, tend to inhibit the development of their own conidia.

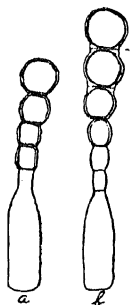


FIG. 13

FIG. 13. The "connective": Diagrammatic representation of two sterigmata with developing chains of conidia; *a*, with outer and inner wall of conidia in actual contact hence no "connective"; *b*, with outer wall partially splitting away from the inner hence showing the connective at *c*.

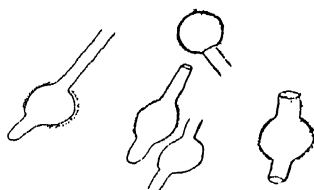


FIG. 14

FIG. 14. Germinating conidia: A rough walled form showing germination by one or two tubes.

Other species shower the nearby areas of culture media with their ripe conidia which quickly cover the whole area with new colonies. In most species the chains of conidia remaining within the confines of the colony refuse to germinate. In some forms, transpired drops are quickly covered with the mycelium of germinated spores, or spores falling to the surface of the substratum, produce mycelia which overgrow the original colony. Attempts to seal and store cultures of such species have failed, if the substratum present retained sufficient moisture to induce germination. Colonies of *P. citrinum* Thom, stored in sealed condition, after a few weeks often show the original colony to be free from ungerminated conidia and to be overgrown with fine mycelium which does not fruit and dies fairly quickly.

*Connective*

Once formed the conidium reaches characteristic shape by enlarging and laying down new walls within the primary wall which is continuous with that of the parent cell (fig. 13.) The presence of vestigial cell walls as a connective between conidia in the chain is common in some species and occasionally seen in others. This appearance referred to as a bridge, disjuncter or connective by various authors was fully described by Thom, 1914, and shown to be merely incidental to the formation of the thickened cell wall of the spore itself leaving occasional gaps between the new wall and the old, when rounding off splits the conidia apart at the septa or ends of each cell.

*Size of conidia*

The measurements of conidia form a part of every species description. These are expressed in microns ( $\mu$ ) either as the diameter or range in measurement of diameter for globose cells or as the long axis by the shorter axis for elliptical cells. In our species, some of which have been kept in culture for more than twenty years, measurements show a fairly consistent range of variation. In making the determinations, we work with an ocular micrometer always present in the field, hence giving a continual check on observations. In mounting portions of a colony for such observations a part selected as typical under a  $10\times$  handlens is removed under the lens from the colony in the petri dish with a sharpened nichrome wire and teased carefully. Where tube cultures must be used the area to be sampled with a hooked transfer needle is first carefully examined with the handlens, and the sampling is usually repeated at least once to insure representative material. In the ordinary working field in such a preparation, there are hundreds of conidia in which the range of measurements is carefully observed and recorded, together with the limits most frequently seen. The results have been expressed in various ways commonly as a range of measurement, in some species as about 2, 2.5,  $3\mu$ , or some such figure, allowing a range of 0.5 micron in either direction. Few lots of conidia fail to show considerable range in diameter, hence figures to the tenth of a micron mean little unless the variation is indicated or implied.

Differences between the measurements of conidia as reported by different workers are striking. Maire (1926) has recently analyzed variations in the reports of spore measurements for species of fleshy fungi as involving a series of causes (1) study of different species under the same name, (2) examination of spores in fluids which shrink or swell the spore, (3)

the measurement of immature spores, (4) actual errors in observation, (5) errors in standardization of the micrometers used. As a result of consistent practices all of the measurements of certain workers tend to be large, those of others to be low. The same conditions apply in studying molds. In the study of *Penicillia* the occasional presence of a greatly enlarged conidium among hundreds reasonably uniform in size scarcely warrants increasing the range of measurements reported to include spores of twice the usual diameter. Such observations call for further study, however, to account for the contrast in size. In some cases these enlarged cells seem to be spores which are in the early stages of germination. Again there are accessory spore forms in certain groups.

#### *Germination of conidia (fig. 14)*

Conidia of any species inoculated into a favorable nutrient germinate in essentially characteristic manner. Some species swell greatly before putting out one or more germ tubes; some merely "round up." Germ tubes may be put out from special areas as in *Scopulariopsis* or from any point on the surface of the swollen conidium. The number and arrangement is characteristic in some species and apparently not characteristic in other species.

#### *Colony growth*

The germ tubes quickly elongate, divide into cells by septation and branch extensively to form closely woven networks, felts or mycelia. In a favorable nutrient evenly distributed as in a petri dish the resulting colonies are usually approximately circular although marginal areas may be characteristically even, notched or stellate or crenulate on account of the establishment of main radiating hyphae from which the interspaces are filled by branching systems. Muller, working with a strain reported as *P. glaucum*, attributed this radial habit of growth to negative chemotropism toward its own metabolic products which accumulate most rapidly toward the center of the colony. Smith working with a strain of *P. expansum* (our no. 4187) reported that growth in length occurred only at the tips of the hyphae. Cells once formed might throw out branches with new growing points but the cells themselves did not increase in length during the period of observation. Such a theory might account for the plane surfaces of *P. expansum* or *P. roqueforti* but would require adjustment to account for the contorted, cerebriform or radiately wrinkled mycelia of many of the species described by Zaleski.

## ACCESSORY SPORE FORMS

Horne and Williamson (1923) using the generic name *Eidamia* in reporting studies of a strain of the species aggregate, variously named *Paecilomyces* Bainier, *Corollium* Sopp, *Penicillium divaricatum* Thom, have described a type of submerged fruiting branch producing elliptical, terminal cells about twice the size of the ordinary conidia of the species. These cells appeared in Thom's notes as occasional conidia of double size, but otherwise not accounted for at the time of the description of *P. divaricatum*. Manuscript of another strain or, perhaps, species of this same group and fully illustrating the same type of spore has been submitted to us by Kita and Wai of Kyoto Imperial University. Schaposchnikow and Manteifel (1923) describe such terminal cells for a sand colored mold (no. 536), evidently also a strain of this group, *Penicillium arenarium*. Nadson and Jolkevitch (1923) found similar cells in a fungus termed by them *Spicaria purpurogenes*.

More recently, in a strain of the *P. lilacinum* series (see no. 225, Chapter XVII) found in electrotyping solutions containing nickel salts, similar submerged fruiting branches were found and studied. In this *P. lilacinum* strain (no. 4855) careful examination of these fertile branches suggests the hypothesis that the impulse to produce conidia when acting in a submerged hypha in which all parts are in constant contact with the nutrient, produces a transformed branch bearing spores which tend to enlarge, round up as if for germination, and to lose the catenulate arrangement. This may produce a very peculiar spore attached as a single terminal cell or a cluster of cells not recognizable as conidia and without definite arrangement around the tip of the sterigmatic cell.

Long observation of the same spore forms observed by Horne and Williamson in their "*Eidamia*" suggests the application of the same hypothesis in accounting for the forms observed abundantly in that particular series of strains, but which are scarcely, if at all, present upon the submerged mycelium of others nearly related.

## SCLEROTIA

Pseudoparenchymatic masses or sclerotia characteristic in colors as pink, salmon, yellow or gray structures elliptical to globose in form and commonly 100 to 300 $\mu$  in long axis are found abundantly in certain monoverticillate species. Many of these have been studied, held for varying times, and reexamined repeatedly without finding any sign of ascospore production. Some species produce such bodies regularly and abundantly, others occasionally and scantily, but these bodies have not been found in other series.

## PERITHECIA

Perithecia have been reported in three groups of the Penicillia. Brefeld and recently Schwartz report ascospore formation in the Asymetrica, *P. glaucum* and *P. italicum*. Morini reported it for *P. candidum*. Zukal, Wehmer, Dangeard, Klöcker, Thom, Biourge, and Derr report ascospores in various species and strains of the biverticillate section, the *P. luteum* series of forms. Sopp adds to these groups various species in his genus *Acaulium* (*Scopulariopsis* Bainier or *P. brevicaulis* Sacc. and its allies).

Brefeld's studies presented a form in which pseudo-parenchymatic structures apparently sclerotia developed ascogenous central areas during a long ripening period. In the *P. luteum* series the ascogenous mass is quickly developed and surrounded only by a loose mass of hyphae scarcely developing a true perithecial wall. Again these two groups differ markedly in the type of ascospore presented. Brefeld figured disk or lens-shaped ascospores consisting of two halves or valves as in ascogenous *Aspergilli*, each with a membrane or frill making a double equatorial band, and bearing echinulations on the surfaces of the valves. These ascospores are figured in germination with the valves separated and showing upon the opposite sides of the germ tube.

In contrast to this type of ascospore, *P. luteum* produces an oblong or elliptical spore variously echinulated, marked or bearing a spiral frill, but lacking the valve-like structure with its phenomena of germination. Sopp's figures and descriptions show heavy walled perithecia in some drawings including an ostiole, but lack details for further discussion.

Derr in his recent paper reports members of the *P. luteum* group as heterothallic together with his determinations as to the sexual status of various forms under investigation by him, but gives no morphological or cytological data. He identifies one of these strains with *P. vermiculatum* Dangeard, however, thus making Dangeard's elaborate cytological study of that species available as applying to a form now known, but hitherto unrecognizable for lack of data as to its culture and general morphology.

Each of these reports has added to our list of ascosporic forms, but none of them has carried us far beyond Wehmer's analysis (1894, *Hedwigia*) which was about as follows: Most of the described species produce conidiophores only, a few produce ascospores, with various accessory structures; (1) in filamentous masses without differentiated wall as in *P. luteum* and its allies; (2) in definite perithecia with soft thin but definite walls as in *P. insigne* Winter or Schröter (?); (3) within sclerotium-like

masses which ripen slowly as in *P. glaucum* of Brefeld. Sopp added his perithecia in Acaulium with their heavy walls and ostioles.

Bezssonof studying the molds of concentrated sugar solutions, reported (1918) that *P. glaucum* produced perithecia when grown in concentrated sugar solution.

The data given were not convincing but rather indicative of contamination of the cultures with *Aspergillus*.

Derox's studies have now pointed out the possibilities open to the student of sexuality in the *Penicillia*. None of them have gone far enough yet to offer a sound basis for giving the various sections of this group their proper places among the Ascomycetes. The work thus far indicates, however, that when perithecium formation is dependent on the union of haplonts of heterothallic series these haplonts will be found to show close relationship in colony and conidiophore characters. A scheme of classifying these conidial forms which will bring together the related strains into recognizable groups will simplify the task of finding the proper organisms for such study. The task of testing these organisms strain by strain belongs to the specialist in that field. Meanwhile the student of the activities of *Penicillia* as they are encountered in nature, needs a means of identifying conidial forms which will continue to constitute 99 per cent or more of the organisms which appear in his cultures.

#### CYTOLOGY

Gueguen in 1899 reported work upon the cytology of *P. glaucum*. Cultures upon moist bread were taken as normal. Best results were obtained without fixation. The conidial wall was comparatively thick, one-fifth of the total diameter, and showed in optical section alternate areas thin and reinforced, hence differently staining. In the germinating spore the old wall was seen and figured on the remains of the swollen cell in contrast to the thin wall of the growing hypha. He depended upon intravital staining with gentian violet or dahlia in very dilute solutions. The vegetative cells were multinucleate. The cells of the conidiophore were not different in structure from those of the vegetative hyphae. The branches and metulae were also multinucleate. He figured the young sterigma as at first several nucleate, the actual number being variable, then reduced (fusion is suggested) to two, one of them centrally located, the other at the point of conidium formation. Although two nuclei are figured in sterigma and conidium in the process of separation, he reported only one nucleus in the conidium where separa-



tion was complete. His inference was that before the wall was laid down, one of the two nuclei in the developing conidium migrated back to the apical position in the sterigma to become the nucleus of the next conidium and to be replaced at the apex of the sterigma by another produced by division of the central nucleus. Multinucleate conditions are resumed in the process of germination. Gueguen's discussion of the perithecium and ascus formation lacks figures and the definiteness necessary to add anything to our information. Dangeard's work has been discussed in connection with the historical considerations in Chapter II.

#### MUTATION AND VARIATION

No one can study hundreds of isolations of strains belonging to such groups as *P. expansum*, *P. roqueforti*, *P. chrysogenum* or *P. digitatum* without being impressed with the variability in color of conidial areas, in colors produced in the substratum, and differences in the amount and nature of the mycelial mass. Some of these differences are manifestly the response of that particular culture to the environmental conditions presented, without representing real or permanent differences in the organisms themselves. Other strains have an individuality consisting of minor differences in appearance, reaction or habit of growth, through years of propagation in the laboratory, but when subjected to analysis in seeking a basis for description can only be interpreted as consisting of quantitative differences in characters well established qualitatively for the species. Again, *P. digitatum* var. *californicum*, discovered and sent in by Fawcett, presents a strain lacking entirely the characteristic color of the species yet maintaining the well-known morphology of *P. digitatum* and its specific ability to attack citrus fruits. Such a difference is a valid basis for separation and probably represents a suppression of the factor for inheritance of color. Without debating whether the term mutation, saltation or some other phrase is correctly applicable, the evidence of relationship between these forms is not to be ignored.

The mechanism of variation in *Penicillium* has been studied by Waterman, Haenicke and others. The constant occurrence of anastomoses between the cells of adjacent hyphae in the mycelium and even between conidiophores when packed closely together has been emphasized by Brierley (personal discussion) as bringing in endless cytological possibilities. Practically all of our colonies grow not from one but from many spores and hence present many mycelia which blend and certainly in some cases anastomose. The colony as we have it described is thus a

composite in which the effect of anastomosis remains unanalyzed. The great number of closely related forms encountered suggest great possibilities in the study of pedigreed cultures as establishing the limits of variability within the species and perhaps explaining relationships within these series.

Waterman has suggested inhibition in the development of *P. glaucum* as a cause of mutation. He supports this conclusion by experiments in which mutations were apparently produced by poisons such as copper sulfate and boric acid, narcotics as para hydroxy benzoic and salicylic acids, culture medium with galactose or a polysaccharide containing the galactose group as a sole source of carbon and culture media with glutaric, levo-tartaric, meso-tartaric acids or rhamnose as sources of carbon.

#### *Yeast-like structures*

Fuchs (1926) reports and figures yeast-like cells as produced by his strain of *P. glaucum*. Unfortunately he did not describe his species in terms adequate for identification.

#### *Polymorphism*

The changes in morphology and physiological reaction already discussed have been mostly in response to environment either in the form of composition of the nutrient medium or to combinations involving such factors as conditions of humidity, temperature, pressure, gas ratios, competition of other species, or proximity of other colonies of the same species.

When a favorable environment in the form of adequate nutrients and growing conditions has been rigorously standardized, most of the species of *Penicillium* have been found to maintain unchanged morphology and reasonably stable physiological response. Nevertheless the idea of polymorphism in the organisms themselves which pervaded much of the earlier literature, still persists, and citations from some of this literature are still encountered.

Gueguen (1898, p. 22) discussed the variability of *P. glaucum* in terms which we know now indicate the impurity of his cultures rather than the responses of his organism. Gueguen's results can not therefore be applied to any species from his use of the name alone and unless internal evidence can be found to identify his organisms the data given are useless. How close this work may be related to that of Dangeard is problematical.

## STANDARDIZED DESCRIPTIONS

The diagnoses of *Penicillia* in the literature present many different descriptive terms and a varying arrangement of the data. Frequently more valuable hints leading to identification are found in the author's vernacular notes than in his Latin diagnostic paragraph. The usages of the author can often be best determined by studying his description with his figures. In collecting the mass of information published, we have fixed a general type of description, adopted a series of standardized terms for species already described and then rewritten the available information species by species in our own terms. The citation of the exact place of original publication is given after each species, for the worker desiring to consult such originals. It is believed that the entire recasting of the descriptions in the light of our own studies, and using a single set of descriptive and morphological terms will serve the user of this book, better than a literal translation of the original which for most species is present in our laboratory notes as either a copy or a photostat.

This standardized description specifies as closely as possible the substratum used, the color and color changes of the colony, colors in reverse and substratum, the texture and appearance of the colony, its marginal features, zonation if present, odor, transpired drops; the conidiophore, its measurement, origin and arrangement, the penicillus with its branching system, metulae, sterigmata, conidial chains and conidia. Accessory spore forms, sclerotia, and perithecia, if present, are discussed together with conspicuous features of metabolism when known.

## PLATES AND FIGURES

Great difficulties are encountered in the interpretation of the illustrations which accompany the descriptions of *Penicillia*. Figures suggestive of the penicillate type of conidium production are recognizable in the works of Micheli, Bulliard and other early mycologists. If such figures are based upon the exceptional species whose relations to a particular substratum make a characteristic picture, we may establish a presumption of identity as in part of the figures of *Coremium vulgare* in Corda's *Prachtflora*. If the general features of a conidial apparatus only are presented, identification is rarely possible. Authors with a selection of a small number of divergent types in hand felt only the need of such contrasts as would separate the forms which they had. With hundreds of cultures and the accumulated literature, sharper lines of separation become necessary.

Another danger lies in the over-emphasis of the unusual or bizarre structures occasionally encountered. This illustrated by Thom's figure (1910) 26b of *P. stoloniferum* which while produced more or less regularly by this and related species upon certain gelatine media is made to overshadow the *P. brevi-compactum* type of penicillus so characteristic of this form and seized upon correctly by Dierckx. Another illustration is the vesiculose mycelium described by Bainier for *P. vesiculosum* which we finally found fairly convincingly in a variant or, perhaps, pathologic culture of *P. roqueforti*.

In an elaborate study of a species, the range of variation in structure is important. In seeking a basis for helping others determine a particular species of *Penicillium*, details which are usual in the organization of the penicillus, associated with habit drawings and sketches which give the appearance of the usual form of penicillus and its relation to the mycelium with its usual variations are far more significant. Detail drawings to be useful must show a clear cut knowledge of the cellular history involved in the formation of the penicillus, the position and character of the cell walls, and the shape and markings of the various elements. The unusual morphological response of a particular organism to a particular environment is not to be overlooked, but the unusual must be scrutinized and shown to be a consistent response to conditions before it should be included in either description or illustration.

Too much emphasis can be placed upon exact measurements and numbers. If species could be found in which branches, metulae or sterigmata were borne in three's, four's, or other fixed numbers, it would be convenient, but these are not found. A description specifying the numbers of elements in the group must be interpreted as correct, perhaps, to a particular fruiting mass studied and described or drawn, but only incidentally true to that preparation.

Such considerations emphasize the necessity of the study of the growing colony by direct observation in the petri dish using the best objectives available for the purpose. In this way the worker obtains a sound conception of the organization of the colony, the origin and course of the conidiophores, the origin and development of the penicillus and the whole mass effect of the conidial fructification. This essential conception can not be obtained from material preserved in alcohol, nor from examination of slide mounts of a few hyphae and conidiophores with high magnifications only.

## DIAGRAMMATIC RADIAL SECTIONS

In seeking a method of illustrating the character of colony growth in petri dishes, V-shaped sections from margin to center of typical colonies were removed and studied under the hand-lens in the same field as an accurate millimeter rule. The findings have been combined with the measurements of more delicate structures made under the compound microscope. Diagrammatic sketches have been constructed for such radial sections beginning at the outer edge of growth and running inward radially far enough to exhibit the significant relation of substratum, submerged mycelium, aerial hyphae, conidiophores and where significant penicilli. Lines are arbitrarily used for hyphae and a terminal cluster of diverging lines for the routine representation of the presence of penicilli. In certain species forming long columns of conidia, such columns are represented by parallel dotted lines. A similar representation of colony structure was given by Ezekiel in studying certain Sclerotinias.

## FRAGMENTARY DESCRIPTIONS

Many fragmentary discussions of *Penicillia* are found in the literature. Saccardo (in the *Sylloge*, 20: 282, 1911) cites a number of such instances. The data necessary for identifying members of the genus have already been discussed. It is, as a rule, impossible to identify a *Penicillium* from a partial description or a figure alone. Except in unique forms with some special character, a description fragmentary and unsatisfactory to the describer can rarely be identified by another. Biourge has reproduced a numbered series of species from Dierckx's notes. It is scarcely probable that other workers without the fuller notes and colored plates left by Dierckx can hope to identify these forms. Many of the taxonomic references to *Penicillia* must, therefore, be discarded along with many discussions of unnamed species.

## CHAPTER VII

### PHYSIOLOGICAL ACTIVITIES

Physiological activities in the sense used here comprise those activities which are defined in the terms of the culture laboratory rather than by quantitative chemical analysis (see Chapter VIII). In this sense the fundamental relations of the organism to its environment and those responses which are defined in descriptive terms rather than in measurements are included.

As encountered in nature, *Penicillia* are agents primarily active in the decomposition of organic matter. Plant products rather than animal products are most commonly attacked. Sugars, starches and related substances are especially favorable to *Penicillia*. Sometimes they appear as wound parasites of plant tissue in conditions of low metabolic activities such as over ripe fruits, seeds and stored bulbs. Animal products are not exempt from attack, meat, milk, leather, even bird's eggs in the museum suffer from their activities. Their attacks upon man and other animals form a separate Chapter (X). The amount of nutrient necessary to support a colony of *Penicillium* is so small that it commonly escapes washing processes and may escape the ordinary routine chemical or physical examinations.

### RELATION TO OTHER ORGANISMS

#### *Saprophytes or parasites*

*Penicillia* are usually saprophytes although host indexes record a fairly long list of species as associated with specific substrata or host plants. Careful analysis of original sources discloses few evidences of real parasitism. Rots or spoilage attributed to *Penicillia* may be due to several causes; contamination of broken or wounded tissues by some species of this group along with other microorganisms furnishes a favorable opportunity for mold development. This is usually called wound parasitism. Another type of rot is the destruction of ripe or overripe fruit in which metabolic activity in the host tissue is very feeble. Again in stored products—seeds, fruits, roots, tubers and bulbs—a dormant period has reduced the resistance of host tissues. Only a few of these products may be regarded as actively parasitized. In most cases the

species involved is some cosmopolitan organism whose presence in the host is incidental rather than a specific adaptation to environment. More specific relationships occur in a few cases and certain of these cases will be discussed in the paragraphs which follow.

*Host index.* Compilation of a host index from such published findings was begun but abandoned because scrutiny of the species determinations found showed that such an index would be utterly misleading if references in it to species were to be quoted as valid, and equally worthless if such citation were replaced by the term "*Penicillium* sp." Such references are frequent in the new edition of Farlow and Seymour's Host Index, which has been recently published. The presence of colonies of *Penicillium* in the situations reported is not questioned but the validity of the determination of species is usually doubtful, and barring the occasional case, the organism was incidental to environmental conditions rather than significant as a parasite or a factor in a specific fermentation or decomposition.

Certain more or less specific reports of pathogenic activity will be presented.

### *Parasites*

*Penicillia* are reported among possible parasites of the Irish potato by Edson and by Shapovalov, among possible parasites of corn by Durrell, and of wheat by Beckwith. Nordhausen placed *Penicillium* spores in a wound in a leaf and observed growth but no invasion of uninjured cells. Their ability to penetrate cell walls was discussed by Young.

Miss Johann, working at Madison, Wisconsin, has isolated a strain close to, if not identical, with *P. oxalicum* of Currie and Thom which seems to be parasitic to corn seedlings. It is entirely possible that other species may be found to have parasitic phases. Hoffer reported *P. expansum* as present in seed corn.

Neuwirth in Czechoslovakia in 1924 discussed *Penicillium* as a rot of beet roots. We have studied a strain of the *P. luteum-purpurogenum* group (Clency-Shear 112819-1-B) isolated from sugar beet in 1920. This strain produced long coremia suggestive of *P. duclauxii* but differed in details of structure. Poole sent in another member of this group found upon rotting sweet potato.

*Penicillium* as a bacteriophage. Gratia and Dath record that their strain of *P. glaucum* destroyed *B. anthracis* in cultures.

*Serological.* Etienne (1920) reported spore agglutination with *Scopulariopsis blochii*.

Arnaudi has recently studied the agglutination reactions of the molds of Gorgonzola cheese. Such tests were found a fairly efficient means of separating this series of the *P. roqueforti* group which are scarcely separable by morphological means.

#### RELATIONS TO ENVIRONMENTS

##### *Occurrence*

The universal distribution of species of *Penicillium* has been already noted together with the terms blue mold and green mold variously and indiscriminately applied primarily to *Penicillium* although species of *Trichoderma* and even *Cladosporium* are often included. So abundant and so universally distributed are the conidia of the various species that few natural environments habitable by man fail to be well inoculated with them. The development of moldy areas or moldiness from *Penicillium* is not an evidence of fresh infection but an evidence that conditions of the environment are favorable to mold growth. These conditions are essential nutrients, available moisture and favorable temperatures.

##### *Relations to air*

*Penicillia* are mostly obligate aerobes. Green fruiting areas, for the most part, cover surfaces exposed to the air, or line openings through which air is fully supplied. A few species fruit more or less normally in spaces in which free oxygen is greatly reduced from that of the normal mixture of the atmosphere. Thom and Currie showed that *P. roqueforti* would grow in openings with as little as 5 per cent of free oxygen. Roquefort cheeses when cut frequently show only mold which is pale yellow in color but which turns green in a few minutes upon exposure to the air. Occasional colonies of mold are found in cans of food in which oxygen pressure must have been low.

Mycelium, while massed upon, at, or just under the surface of the natural substratum, sometimes penetrates more or less deeply. Some species appear to be restricted to an outer layer a few millimeters in thickness. Others penetrate for considerable distances but with progressive reduction in mass of mold hyphae. Slow growth under unfavorable conditions is observed in the development of abnormal mycelia among particles fairly deeply in the soil; in the formation of curious masses in sealed bottles and cans (see "bottle imps" described in a succeeding paragraph). Hyphae of mold in solutions, or deeply imbedded usually show marked divergence in structure—slender, or irregular,



zig-zag hyphae among soil particles (Thom and Church), abnormal clawlike growths about sugar crystals, alternate enlarged cells and cells reduced to tenuous tubes in such fatty masses as butter, all sorts of irregular cells in acid or alkaline or concentrated substrata, or developing in the laboratory substrata as the culture progressively dries.

*Vacuum.* Absence of free oxygen practically stops the growth of *Penicillium*. This may be accomplished by high vacuum, by the substitution of an inert gas for the natural mixture of the atmosphere, or by the reduction of the oxygen present through its use in metabolism. Slow development of vegetative hyphae frequently takes place under conditions supposed by the worker to be vacuum but probably very low oxygen tension, but such development without free oxygen is doubtful.

Thom placed a crate of about thirty test tubes inoculated with different species of molds mostly *Penicillia* in a Novy jar. The crate of tubes filled most of the free space in the jar. The jar was sealed and let stand for one week then opened and examined. The colonies (except for *P. roqueforti*) as recorded showed inhibition at about the third day stage of development. When let stand in the room growth was renewed and quickly became normal. Apparently exhaustion of free oxygen or inhibition by some toxic material reached the inhibiting stage in about three days.

In connection with representative studies Kostychev and Afanassjew (Jahr. f. wiss. Bot., 60, 628, 1921), have carried out some experiments on the effect of the absence of oxygen upon *P. glaucum*. They report that, while *P. glaucum* under normal conditions develops in considerable concentrations of organic acids, in the absence of oxygen small traces of acid prove decidedly toxic to the organism. Both *Aspergillus niger* and *Penicillium glaucum* produce ethanol from alkaline sugar solutions under anaerobic conditions; if the solution has an acid reaction *P. glaucum* forms only traces of ethanol. The *P. glaucum* when immersed in sugar solutions with neutral reactions formed fair amounts of ethanol.

*Oxygen pressure.* Karsner and Saphir using "*P. glaucum*" changed the concentration of oxygen present during the incubation period by increases of oxygen up to 50 per cent or even 99 per cent of the mixture without finding inhibition. May and Herrick in unpublished work found an atmosphere of oxygen prevented aerial growth of (no. 2670) their gluconic acid *Penicillium* although some submerged mycelium developed.

*Nitrogen fixation.* Duggar and Davis after many experiments suggest that *Penicillium digitatum* and *P. expansum* as well as some other fungi may fix minute amounts of nitrogen. Ternetz reached similar conclusions in 1907. The amount fixed by these species was regarded by them as negligible however. Pennington (1911) found no evidence of such fixation.

### *Relation to heat*

The majority of the species of *Penicillium* grow best at temperatures below 30°C. Zaleski regarded 22°C., as the most favorable incubation point. Biourge preferred 17 to 19°C. Thom found little inhibition below 30°C. or even 33° or 34°C., but many species which were inhibited at 37°C. McCulloch and Thom obtained an entirely different picture with *P. gladioli* below 15°C., than above 25°C. Fawcett and Barger measured the rate of growth of *P. italicum* and *P. digitatum* in oranges finding the most rapid action between 66.8°F., (19° and 27°C. and 80.5°F., with almost no activity at 90°F. (32°C.+), and much reduced activity at 50°F. (10°C). Recent studies in our own laboratories have given added importance to the range of temperature in the incubator. A number of Biourge's types and certain of Zaleski's types failed to grow normally at 30°C. or above. It becomes therefore increasingly necessary to specify the conditions of incubation in every study of mold action.

Wiesner (1873) reported the optimum temperature for "*P. glaucum*" as 22°C. although mycelium increased continuously up to 26°C., from which point loss of vigor was discontinuous up to a maximum temperature tolerated at 42 or 43°C.

Sartory found the optimum for *P. gratioti* at 34° to 35°C., but growth continued up to 49° or 50°C.

*Pasteurization, cooking.* Conidia of *Penicillia* suspended in milk are readily killed by heat. Thom and Ayers tested thirty-two named species and eight unnamed strains of *Penicillia* suspended in milk by subjecting them for a period of thirty seconds to a series of temperatures. None survived at 175°F. (79.5°C.), two doubtful survivals were reported at 165°F. (73.9°C.), two different species survived at 155°F. (68.3°C.); many species grew after exposure to 145°F. (62.8°C). for thirty seconds. Any cooking process therefore recognized in household practice will destroy these spores. Using the same series of organisms and dry heat, four species grew after thirty minutes at 230°F. (110°C.); no species survived 250°F. (121.1°C.) for the same period.

*Relation to hydrogen-ion concentration*

Kouznetsoff (1925) working with "*Citromyces glaber*" (see No. 79) reported his organism as finding its optimum at pH 5.5 with range of growth from 8.7 to 2.7. Webb studying the germination of conidia found increasing acidities up to pH 4 or even to 3, favorable to germination where alkaline conditions were distinctly unfavorable. He used *P. cyclopium* and *P. italicum* together with species of other genera.

Gustafson (1920) found that the rate of respiration of *P. chrysogenum* was not affected by a change in hydrogen ion concentration from pH. 4 to pH 8. At pH 8.8 the rate fell to 60 per cent of the normal while at pH 2.6 an increase was noted followed by a gradual return to normal. In general the oxygen consumption was found to increase in an acid and to decrease in an alkaline solution. He further (1920) reported that a solution of dextrose and hydrogen peroxide behaved like *P. chrysogenum* when cultured on dextrose solution in that an increased quantity of carbon dioxide was produced when the solution was acid but less when the solution was alkaline.

*Relation to light*

There is very little definite information as to the effects of light upon species of *Penicillium*. Great difficulty is ordinarily encountered in separating the effects of light from other physiological factors in the same experiments.

The exposure of petri dishes inoculated with conidia to sunlight in working rooms for several days has seemed to destroy the viability of the spores used. Too many variable and uncontrolled factors were acting to render the results significant. Special studies with particular types of radiation are much more satisfactory.

*Ultra-violet radiation.* Fulton and Coblenz studied the effect of ultra-violet radiation as a means of destroying mold spores especially *Penicillium digitatum* and *P. expansum* on the surfaces of citrus fruits. They used a 110-volt quartz burner having a mercury cathode and a tungsten anode normally operated on 320 watts (80 volts, 4 amperes). They found such radiation efficient in killing the conidia when the rays could be directly applied. "An exposure of five seconds at six inches was sufficient to kill at the rate of 907 out of 1000 spores of *P. digitatum*;" "with an exposure of 45 seconds the rate of killing was 998 out of 1000." From the practical standpoint the failure of the rays to penetrate hence to destroy mycelia which had already invaded the tissue, and the difficulty of producing adequate exposure of the entire surfaces of the fruits

render the method one of very limited usefulness. Its value would lie primarily in its application to special problems in which the entire substratum could be readily exposed to radiation.

Irradiation of lipid containing substrata for short periods was found by von Euler (1925) to stimulate the growth of *P. glaucum*; when longer periods were used growth was retarded.

DeGraaf using a quartz mercury vapor lamp found the conidia all dead after five minutes exposure at 30 cm. Spores suspended in liquid fats were not killed even in thin layers.

### *Moisture relations*

If we assume fairly general inoculation with spores, and if the necessary nutrients, gas ratios and favorable temperatures are present, absence or development of *Penicillium* is determined by the availability of the moisture necessary for growth. To be available for growth the water must not only be present as shown by analysis but must carry the soluble constituents of the substratum at such dilution that part of the water itself is obtainable by the mold hyphae. This concentration of solutes varies with the substratum and the species of *Penicillium*. Some species have been shown to grow in solutions of sodium chloride at 10 per cent or higher (see *P. roqueforti*). Many species will grow in sucrose solutions up to 20 per cent; some grow in much more concentrated sugar solutions. Species by species osmotic pressures, however, can be determined at which growth ceases. As a result, moldiness will develop in different substrata at very different water percentages and will present aspects differing with the species of *Penicillium* present.

Pfeffer in 1889 reported the following as the limits of concentration tolerated by *P. glaucum*, glucose 55 per cent, glycerine 43 per cent, sodium nitrate 21 per cent, calcium chloride 18 per cent, sodium chloride 17 per cent.

Grove found 65 per cent of sucrose necessary to prevent the growth of his strain of *P. glaucum* in jams and jellies. Beauverie (1901) reported that the greatly increased osmotic pressures reduce the aerial growth of mycelium in contrast to the submerged portion of the mycelium.

*Distilled water.* The impurities of laboratory distilled water are often emphasized. Tubes of water (prepared by Thom) from the block tin still then in use in the chemical laboratory of the Storrs Agricultural Experiment Station were inoculated with spores of forty-four strains of *Penicillium*. The conidia of seven of these strains germinated to

sufficient extent for growth to be seen under the hand-lens by careful examination of the under side of the meniscus in the tube. No further development occurred.

### *Metabolic products*

Nikitinsky (1914) analyzed the effects of the products of metabolism upon certain species. Frequent references throughout this book, emphasize the changes in colony color due to the change in hydrogen-ion concentration of the substratum during colony growth. Microscopic studies of old colonies show swollen or vesicular cells of many forms with or without regularity attributable to the conditions of growth. The reflex effect of the changes in the substratum upon the colony must not be overlooked in interpreting observations of mold cultures.

*"Staling" effects.* Colonies of the same or different species growing on the same substratum may or may not exert marked influence on each other. Various authors (Pratt, Harder, Zellar and Schmitz) have discussed these phenomena using *Penicillia* among the fungi tested. There is no question as to the observed effects in inhibition or stimulation of growth, changes in colony color, color in the substratum (already discussed in Chapter V).

*Vitamins, etc.* Gay (1921) reported that accessory substances such as vitamins were not necessary for *Penicillium glaucum*. Many difficulties are involved in establishing this as a general or specific character. The amounts of some of these substances required for a normal colony of *Penicillium* are so minute that our ordinary laboratory precautions in cultivating molds will not exclude them as contaminations. There are certain species of the group which tend to die out in transfers and which no one has successfully maintained for a long period, whereas other forms are kept readily; some species in our collection seem to be unchanged after twenty-five years of continuous transfer. Lack of essential nutrients has been assumed as the cause of these losses which, so far, have not been successfully combated.

## RELATIONS TO ANTISEPTIC AND PRESERVATIVE MEASURES

### *Disinfectants and preservatives against Penicillia*

Many efforts have been made to find harmless antiseptics which may be used to prevent the growth of molds such as *Penicillium* in food-stuffs. Salt (NaCl) is effective against most species when its concentration in the moisture present is 10 per cent or over; some species are more

tolerant. Most spices (Bachmann) are feeble inhibitors of mold when effective at all; sugar solutions are effective only as they pass the tolerance of various species to osmotic pressure; such acids as acetic, lactic, and citric are effective but require concentrations beyond the toleration of the human palate. Various poisonous substances are effective against *Penicillium* but as a rule the percentage required is beyond the safety line for human use. Perry and Beal found *P. glaucum* (? what species) to grow in the presence of alcohol up to 8 per cent and to remain viable up to 14 per cent; in sodium salicylate, growth ceased at 3 per cent and the spores were killed at 5 per cent; in sodium benzoate visible growth ceased at 0.25 per cent; formaldehyde inhibited growth at 0.25 per cent and destroyed the mold at 0.4 per cent. Such figures leave no opening for the preservation of food by these substances without danger to the consumer.

#### *Coefficient of toxicity*

Le Renard sought to establish a coefficient of toxicity using "*P. glaucum*" (without data as to what actual species) as a test organism, a standard nutrient and a series of toxic agents. From the results given it would appear that such a relative figure could be readily established for any particular nutrient substratum and test organism but would need to be reestablished with each change of nutrient or test species.

#### *Effect of Acids*

Free acetic acid according to Reichel (1910) has a restraining effect upon *P. glaucum* in a basal Raulin's solution to which additional test substances were added including glucose, cane sugar, tartaric acid, and aluminum acetate.

*Boric acid.* Edmondson, Thom and Giltner tested eight species of *Penicillium* in tubes of dextrose agar to which a canning powder containing 95 per cent boric acid and 5 per cent common salt was added in percentages of 0.63, 0.724, and 0.87. In these experiments one species, *P. camemberti*, produced normal colonies in all tubes; *P. spinulosum*, *P. expansum* and *P. divaricatum* produced more or less growth without characteristic fruiting. The other species failed to germinate. In another experiment a small colony of *P. roqueforti* appeared upon a cherry in a can supposed to contain about 0.6 per cent of this powder.

According to Böeseken and Waterman, boric acid in a concentration of 0.006 per cent has a distinct inhibiting effect on the development of

*P. glaucum* which is apparently connected in some way with the organic materials present in the nutrient solutions.

Oxalic acid in 0.5 per cent strength was added (by Thom) to Czapek's solution and the tubes inoculated with 38 species. Eleven species produced normal colonies among them *P. camemberti*, *P. expansum*, *Paecilomyces varioti*, *P. funiculosum* and *P. spinulosum*. The remainder of the series showed slight or negligible growth.

Tartaric acid in rather high concentrations (35 cc. N/10 NaOH required to neutralize 5 cc.) inhibited thirty-two out of forty-one species tested by Thom. Among the forms found tolerant were *P. stoloniferum* (See no. 185), *P. granulatum*, *P. spinulosum*, and certain related strains of these series.

#### *Miscellaneous antiseptic substances*

Young, seeking seed disinfectants, found chloramine T at 3 per cent between 95 per cent and 100 per cent effective; uspuline at 0.5 to 1 per cent usually 100 per cent effective.

In a study of the influence of a large number of hydroxy derivatives of benzene on the development of *P. glaucum*, Böeseken and Waterman (1912) found that the greater the solubility of a compound in oil the greater was its growth preventive effect.

Sodium silico-fluoride in 0.25 per cent solution has been recommended by various workers in the rubber industry to aid in keeping out molds, among which they name *Penicillia*. Paranitrophenol, used as a soaking solution in dilutions to 0.3 to 0.15 per cent for a three hour period (Stevens), was also effective in preventing spotting of rubber.

In combating fungus activity on paper pulp (in which *Penicillia* were included) the Forest products workers of the United States Department of Agriculture (Kress et. al.) tabulated their studies with one hundred and twelve different chemicals and obtained their best results with borax, boric acid, a solution of naphthalene in crude cymene, sodium fluoride, sodium dinitrophenolate, and sodium dichromate.

*Spices.* Bachmann tested a species of *Penicillium* for sensitiveness to the essential oils in ground cloves, cinnamon and allspice. Cloves in the proportion of one part to twenty-five of agar medium inhibited this particular species of *Penicillium*. Cinnamon inhibited in the proportions of one to a hundred, lower dilutions not being tried. Allspice inhibited in the proportions of one to fifty but not one to a hundred. In our hands this *Penicillium* was a delicate species which died in culture before identification was completed.

TABLE 2

*Effect of certain acids and drugs upon "P. glaucum"*

CONCENTRATION				
	<i>per cent</i>			
Mercuric iodide...	0.0005	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Mercuric chloride.	0.002	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Silver nitrate.....	0.003-0.005	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Iodoform.....	0.01	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Beta naphthol....	0.02	Gueguen, Bul. Soc.	Myc.	
		France 15, 22, 1899		
Salicylic acid.....	0.1	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Guaiacol.....	0.05	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Phenol.....	0.1	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Copper sulfate....	0.1	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Resorcinol.....	2.0	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Thymol	Inhibits activity at saturation	Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Menthol		Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Camphor		Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Salol		Gueguen, Bul. Soc.	Myc.	
		France, 15, 22, 1899		
Boric acid.....	0.006	Böseseken and Waterman		
Sodium bisulfite...	0.25	Perry and Beal		
Formaldehyde.....	0.25-0.4	Perry and Beal		
Sodium salicylate.	3.0	Perry and Beal		
Salicylic acid.....	0.14	Sabalitscha and Dietrich, Dis-		
		infektion, II: 67 (1926)		
Sodium salicylate.....	1.5	Sabalitscha and Dietrich, Dis-		
		infektion, II: 67 (1926)		
Phenol, Thymol, Carvacrol....	0.014	Sabalitscha and Dietrich, Dis-		
		infektion, II: 67 (1926)		
Pyrocatechol, dimethylether	0.14	Sabalitscha and Dietrich, Dis-		
phenyl acetic acid.....				
		infektion, II: 67 (1926)		



TABLE 2—*Concluded*

	CONCENTRATION	AUTHORITY
	<i>per cent</i>	
Pyrogallol, phloroglucinol, hydroquinone.....	No effect at 1.47 per cent	Sabalitscha and Dietrich, Disinfektion, II: 67 (1926)
Hydrogen peroxide.....	0.05	deGraaf, Nederland. Tijdschr. Hyg., 3, 249, 1928
Quinine.....	0.1–0.3	deGraaf, Nederland. Tijdschr. Hyg., 3, 249, 1928
Eucupine.....	0.03–0.1	deGraaf, Nederland. Tijdschr. Hyg., 3, 249, 1928

Some of the previous discussions together with some material not mentioned are summarized in table 2. Since the conditions under which the various investigators worked were widely variable the data tabulated indicate only in a general way the effectiveness of some of the materials, against some strain designated "*P. glaucum*."

## RELATION TO METALS

*Arsenic*

Gosio first reported that *P. brevicaulis* (Scopulariopsis) when grown upon a substratum containing even a trace of arsenic would evolve arsine. Numerous workers have investigated this characteristic and the possibility of using it as a test for the presence of arsenic in minute amounts. The whole field was carefully reviewed by Huss in 1914. This characteristic was originally believed to be limited to a particular strain identified as *P. brevicaulis* Sacc., but experiments have extended the number of fungi which give this reaction to include all species designated by Bainier's genus *Scopulariopsis* or Sopp's *Acaulium*. Biourge has included the species of *Stemonitis*; Depoorter, in 1921, apparently obtaining his determinations from Biourge, reports *P. divaricatum*<sup>1</sup> Thom as the most active form. Our studies of Biourge's culture indicate that this was from a member of the *S. brevicaulis* group. In his studies Depoorter found the amount of arsenic detectable to be as

<sup>1</sup> Judged by the cultures received from him, Biourge in 1923 did not understand Thom's conception of *P. divaricatum* or the diagnostic qualities which distinguish *Scopulariopsis* of Bainier.

small as 0.00008 per cent in contrast to 0.008 per cent reported in previous work.

Little information is available as to tolerance of arsenic in solutions. One strain (4746) closely related to *P. purpurogenum* was recovered from a solution of 0.5 per cent  $\text{As}_2\text{O}_3$  made up by the pharmacologists of the Bureau of Chemistry. In this solution small mold colonies developed during a standing period of five months.

#### *Calcium*

According to Coupin (1927, vol. 184) calcium appears to be essential for the proper formation of conidia by *P. glaucum*, for when the organism was cultured on a modified Raulin's solution that contained no calcium, a white mycelium was obtained with very slight conidial formation; if 0.05 per cent of calcium as a soluble salt were added abundant mycelial and conidial formation was obtained.

#### *Copper*

Trabut (1895) reported a species *P. cupricum* with red spores as growing upon copper sulphate in fairly strong solutions. De Seynes (1895) however reported that Traub's species was *P. glaucum* and pointed out inaccuracies in the calculation of concentrations used by Traub. Later Gueguen (1899) working with his "*P. glaucum*" reported that his organism grown upon solutions of 1 to 200 copper sulphate gave the reactions reported by Traub, hence decided that a separate species was not justified.

Hattori reported copper-sulphate as acting as a stimulant to *Penicillium* when present at 0.008 per cent concentration.

#### *Gold leaf*

Miyoshi studying the ability of fungi to penetrate membranes found the hyphae of "*P. glaucum*" to branch richly on the surface without forming special haustoria. He reported that the hyphae penetrated his specially prepared gold leaf although he gave no details of his proof but in his later discussion he interpreted the penetration of membranes as due to the secretion of enzymes.

#### *Nickel*

*Penicillium lilacinum* was found in the nickel electrotyping solutions in factory use in the United States and in Canada. Blum and his col-

leagues working at the Bureau of Standards found that the vats of nickel solution developed mycelium of *P. lilacinum* as colorless hyphae floating and submerged thus coming in contact with the surface of the metal plates. Such bits of mycelium lodging on the engraved surface prevents the deposition of nickel at that point and leaves a flaw in the finished product.

### *Selenium*

Nemec and Vaclav reported that very slight dosages of selenium increased the growth of *P. candidum* (probably *P. caseicolum* Bainier) and *P. roqueforti* in a modified Raulin's solution.

## CHAPTER VIII

### BIOCHEMISTRY<sup>1</sup>

References to "*Penicillium* sp.," *P. glaucum*," "*P. crustaceum*," "some" *Penicillium*, just "*Pencillium*" or more rarely some particular species of *Penicillium*, occur hundreds of times in chemical and biochemical discussions of fermentation, decomposition, and related processes, during a period of half a century. Comparatively few of these references to *Penicillium* are accurately enough limited to identify the organism used, today, hence we have made no attempt to refer to all of them. Most of the users of such nomenclature have assumed (without investigation) that the name given was sufficiently distinctive to carry the necessary information to the reader of their discussions.

The existence of great differences between biochemical activities of nearly related molds was not recognized by most of the earlier investigators. To them a green *Penicillium* was closely enough identified by the name *P. glaucum*, without realizing that greenness as a common character of two cultures did not insure that other characters were in any real sense comparable. In other words, greenness has no more significance in predicting biochemical ability of a *Penicillium* than bayness of a horse in judging his speed in a horse race.

Another series of workers made extensive studies of the biochemical activities of particular strains of *Penicillium* without raising any question of the comparative activities of other strains. The papers of Dox, Alsberg and Black, report carefully planned experiments with certain species but offer little or no indication of the activities of species even closely related.

### COMPOSITION OF *PENICILLIUM*

Cramer analysed the conidia of *P. glaucum*, and gave the following figures: Albumins 28.4 per cent, "starches" 17. per cent, alcohol extract 30.4 per cent, ether extract 7.3 per cent, "cellulose" 1.5 per cent, ash 1.9 per cent, unclassified 3.8 per cent. He also (1891) analyzed the mycelium of "*Penicillium*" finding 7.11 per cent of dry substance and 92.89 per cent of water.

Sieber (1881) found 84.7 to 85.7 per cent of moisture in "*Penicillium*."

<sup>1</sup> Prepared by Dr. O. E. May of the Bureau of Chemistry and Soils.

## METABOLISM: SUBSTANCES UTILIZED

The general distribution of the *Penicillia* gives some indication of the widely different substrata from which they can derive the necessary elements for growth and reproduction. Since the introduction of synthetic nutrient media by Pasteur (Compt. Rend., 51: 298-99 (1860)) a large amount of work has been done with a view to defining the elements essential for growth and reproduction of fungi and many of these experiments were carried out with cultures of *Penicillia*. This field is today by no means a closed one and a vast amount of work remains to be done before any semblance of a general correlation of experimental facts can be hoped for.

The early chemical work upon the utilization of carbon compounds was incidental to the widespread interest in the resolution of optical isomers from racemic compounds following Pasteur's studies of this type of isomerism. Pasteur (Compt. Rend., 46: 615 (1858)) first utilized living organisms to obtain an optically active component of a racemic body. The process depends on the ability of the organism to assimilate one of the isomers to a greater extent than the other, thus leaving a preponderance of one of the active forms in the nutrient solution. *Penicillium* "*glaucum*" early came into favor in work of this kind. Pasteur utilized it to obtain levo-tartaric acid from the racemic compound and it was widely used during the thirty years following. In many cases the organism designated *P. glaucum* was undoubtedly a mixed or at best, an ill defined culture, since carefully identified strains were unknown to many of the investigators of that period. Winther summarized the literature dealing with the biological resolution of racemates to 1895. Among the optically active compounds obtained by the action of *P. glaucum* on the solution of the racemic form with added inorganic salts the following are mentioned: Acids—*d*-mandelic, *l*-glyceric, *d*-lactic, *l*-leucine, *l*-tartaric, *l*-glutaminic and *d*-ethoxy succinic; Alcohols—*l*-methyl-ethyl carbinol, *l*-methyl *n*-propyl carbinol, *l*-methyl-butyl carbinol, *d*-methyl *n*-amyl carbinol and *d*-ethyl propyl carbinol. Or, stated otherwise, the optical isomers of the above compounds are better sources of carbon for *P. glaucum* than those named.

The biological method of resolution has been superseded entirely by the more rapid and more satisfactory chemical methods but the question of why and how the space arrangement of the constituents of a molecule governs the assimilability by fungi, of a compound remains of great theoretical interest.

Herzog and Meier in a study of the oxidation of optically active

organic acids by *P. glaucum* found that the ease of oxidation between the isomers varied depending on the acids used. Thus meso tartaric acid was slowly oxidized and *d*-tartaric acid was more easily oxidized than the levo form. With mandelic acid, the levo form was the more readily oxidized. Little difference was observed in the rate of oxidation of the dextro and levo forms of lactic acid.

Dox (1910) has published some experiments regarding the availability to several strains of *Penicillia* of various compounds exhibiting cis-trans isomerism. He used fumaric, maleic, mesaconic (methyl fumaric), citraconic (methyl maleic) and itaconic (methylene succinic) acids. Table 3 taken from his paper gives the results of his experiments.

Since mesaconic and citraconic acids bear the same relation to

TABLE 3

*Availability to several strains of Penicillia of various compounds exhibiting cis-trans isomerism (according to Dox)*

		MESA- CONIC	CITRA- CONIC	
<i>P. camemberti</i> ....		G	0	0
<i>P. roqueforti</i> .....		G	0	0
<i>P. expansum</i> .....	***	G	0	G
<i>P. chrysogenum</i> ...	***	G	0	*
<i>P. purpurogenum</i> .			0	

G indicates germination only, \*slight growth, \*\*fair growth and \*\*\*good normal culture and 0, no growth at all.

each other as regards solubility, configuration and anhydride formation as do fumaric and maleic acids the results obtained with these two compounds were quite unexpected.

Verkade and Sohngen found that *P. glaucum* readily assimilated fumaric, cinnamic, allo-cinnamic, aconitic, oleic and erucic acids while maleic, citraconic, mesaconic, itaconic, isocrotonic, angelic, tiglic and undecenoic acids were not attacked. They concluded that it was inadvisable to distinguish between cis and trans isomers on the basis of their reactivity or inertness toward fungi and that assimilability depends on the molecular configuration of a compound.

Hasselbring (1908) utilized a selected strain of "*P. glaucum*" to make a study of carbon assimilation from ethonal, potassium ethyl sulfate, ethyl nitrate, ethyl acetate, potassium acetate and acetic acid. It was found that ethanol, acetic acid, ethyl acetate and potassium acetate were assimilated. No spore formation occurred with ethanol

as the source of carbon. While ethyl nitrate and potassium ethyl sulfate permitted germination, the organism was unable to utilize their carbon for growth. Acetic acid proved to be toxic in concentrations of 0.016 N but was a good source of carbon in concentrations of 0.008 N.

Coupin (1927) has recently made a report of a series of experiments dealing with the assimilability by *P. glaucum* of various aromatic and aliphatic compounds.

As a basal substratum he employed a liquid with the following composition: water, 2000 cc.; ammonium nitrate, 6 grams; potassium phosphate, 1 gram; potassium chloride, 1 gram; calcium nitrate, 0.60 grams; magnesium sulfate, 0.60 grams; ammonium sulfate, 0.30 grams; zinc sulfate, 0.05 gram; manganese chloride, 0.05 gram; potassium silicate, 0.05 gram; 125 cc. of the solution was used and carbon containing compounds were added to the extent of 0.5 gram, if solid, and three to five drops if liquid and the flasks and their contents sterilized at 120°C. for fifteen minutes and inoculated with conidia grown upon carrot. Test flasks were incubated for ten days. Ability to utilize a particular source of carbon was indicated by abundant mycelium and conidial masses. Among the primary alcohols ethyl alcohol was assimilated but not methyl, propyl, butyl, allyl or hexyl alcohol. In the polyatomic series of alcohols erythritol, mannitol and glycerol were satisfactory sources of carbon but glycol was not. Ether, formaldehyde, chloral and acetone were not utilized.

Fats were all good sources of carbon but difficulty was experienced in obtaining them in pure condition and of incorporating them in the basal nutrient solution. Excellent growths were obtained upon glucose, levulose, galactose, mannose, sucrose, maltose and raffinose and a poorer growth on lactose. Salicin, amygdalin, inulin, dextrin and glycogen were found to be good sources of carbon but cooked starch, gum arabic and cellulose from cotton were not utilized by this *Penicillium*. Malic and succinic acids were assimilated but formic, acetic, butyric, oleic, malonic, glycolic, lactic, tartaric and citric were not. Glycocoll and asparagin gave poor growth. In the aromatic series Coupin reported phenol, picric acid, naphthol, pyrocatechol, pyrogallol, phloroglucin, benzaldehyde, quinine, salicylic acid, camphor, furfurol, quinine, strychnine, nicotine and quinine sulfate as not assimilable. Sterile submerged mycelia were produced with resorcinol and some floating mycelia with some conidia upon hydroquinine. Among the compounds furnishing carbon for the organisms were tannin, gallic and hippuric acids. The failure of such workers to describe their organisms accurately is most regrettable.

Sartory in studying *P. gratioti* found that sugars were available for growth in the following order beginning with the most readily utilized: sucrose, maltose, levulose, lactose, galactose, inulin.

### *Nitrogen metabolism*

Literature concerning the nitrogen metabolism of definite strains of *Penicillium* is scanty. Various investigators have included some "Penicillium" in studies which have dealt primarily with other molds such as *Aspergillus*. Pfeffer made a study of "election" by *P. glaucum* about 1889. Such workers as Klotz conclude that nitrogen sources which are suitable for *Aspergilli*, are satisfactory for growing *Penicillia*. They list among the compounds reported as utilized, nitrates, ammonium salts, peptones, certain amino-acids and alkaloids, when in proper combination with products which furnished the other essential elements in available form. Further studies in the specific ability of the known species to utilize accurately selected nutrients will be needed to put our information in definite terms.

There is a vast accumulation of culture experiences in which unnamed or vaguely defined species of *Penicillium* have been found to thrive upon or refuse, equally vaguely defined substrata. Properly supplied with other nutrients the extracts of meats, eggs, milk, carrots, beans, potatoes, apples, oranges, rice, most cereals, etc., have favored growth. Corn meal without supporting nutrients has been less generally favorable for *Penicillium*.

The makers of synthetic substrata have added asparagin, nitrates, ammonium compounds, amino-acids, amides, etc.; sometimes limiting their molds to a single source of nitrogen; again including both nitrate and ammonium radicals.

We know from such accumulated experience that organic extracts, from meat, potato, cereal, or other products lacking in soluble carbohydrate if supplemented by some sugar or starchy compound produces vigorous growth of most species. Meat alone, gelatine or meat extracts alone produce feeble and often colorless colonies, of most of these species. Such products appear to be unfavorable sources of carbon since the addition of sucrose to those unfavorable nutrients insures prompt and vigorous growth with abundant colored conidial areas. They are therefore favorable sources of nitrogen.

In other words, there is abundant experience with substrata favorable and unfavorable for growth of *Penicillium* but comparatively little exact chemical data as to the availability of definite nitrogen compounds to definite species of *Penicillium*.



No attempt will be made here to synthesize the multitude of references to the growth of *Penicillium*, into a homogeneous analysis of mold metabolism upon this or that compound. A series of paragraphs in approximately alphabetical order to compounds or products discussed, will give an idea of the range of studies reported with various species of the group, and offer references to investigations and bibliographies more extensive than can be given here.

Agar-agar used (by Thom) as 1.5 per cent in distilled water and inoculated with about twenty species, produced only slight growth consisting of a few radiating hyphae across the surface of the substratum with scanty development of conidial masses. The amount of growth produced in these experiments proves nothing except that there were nutrients enough present to support slight colonies which might be added to or taken away from the usual colonies of the species without the change becoming evident. It was concluded that the addition of agar-agar as a means of substituting a solid for a liquid substratum introduced no real error in physiological studies of *Penicillium*.

*Cobaltamin.* Kinosita used a culture designated *P. glaucum* in connection with *Aspergilli* in a study of various cobaltamin salts as sources of nitrogen. The nitrogen content in each case was adjusted to constant value. In all cases growth was much less than for control experiments in which ammonium nitrate and potassium nitrate were used. In the few salts which permitted appreciable growth the mycelium contained a high percentage of carbon.

*Cocconut oil.* Stokoe (Action of Molds on Cocconut Oil. *Biochem. Journ.*, 22, 80-93, 1928) has found that the rancidity of cocconut oil owing to the action of *P. palitans* is caused by the presence of ketones, chiefly methyl amyl, methyl heptyl and methyl nonyl. The characteristic perfume odor of the rancid oil is caused by methyl amyl ketone which is formed in the greatest quantity. The production of the methyl ketones indicates that in this *Penicillium* as well as in animals and man, oxidation of a chain compound takes place at the B carbon atom with the formation of a keto acid, the normal decomposition of which would result in the formation of a fatty acid containing two less carbon atoms and acetic acid. In the case of *P. palitane* the absorption of poisonous fatty acids impedes the normal respiration, bringing about a condition whereby the keto acid is decomposed into a methyl ketone and carbon dioxide. Stokoe holds that only acids up to lauric acids are absorbed and hence ketones of higher molecular weight than methyl nonyl ketone are not formed.

Acklin has studied the formation of methyl ketones by *P. glaucum* from the ammonium salts of the normal monocarboxylic fatty acids from butyric up to and including myristic acid, a range of from 4 to 14 carbon atoms. Butyric and valeric acids showed no ketone formation but with the remainder of the series the production of methyl ketones was demonstrated. Acklin concludes that the reaction proceeds by way of B oxidation thus involving the formation of a keto acid which is further oxidized to the ketone and carbon dioxide, acetic acid and possibly secondary alcohols being also formed. He also investigated some of the conditions governing the formation of methyl propyl ketone from caproic acid by *P. glaucum* and found that the percentage yield of ketone was dependent on the concentration of the acid in the substrata. Higher yields were obtained when the triglyceride of the acid was used. Solutions of this ester buffered to pH 7.6 and 4.2 gave percentage yields of ketone of 35 per cent and 30 per cent respectively while unbuffered solutions with an initial pH of 7.6 gave percentage yields of 48 per cent.

*Cyanamide.* Kappen grew a green *Penicillium*, *P. brevicaulis*, and other fungi upon solutions of cyanamid. His green *Penicillium* and *Cladosporium* grew in solutions up to 0.5 per cent, the others up to 1 per cent. The enzyme found active was an endoenzyme related to but not identical with urease.

*Gold.* Williams grew "*P. glaucum*" in a colloidal gold solution containing tannin or gum arabic and found that gold was taken up and retained in the non-cuticularized walls of the cells.

*Lactic acid.* Lactic acid in tenth normal strength (0.9 per cent) was found by Thom to supply carbon for normal growth of *P. camemberti*, *P. caseicolum*, *P. bifforme*, *P. commune*, *P. duclauxi* and one strain of the *P. chrysogenum* series, all of which except *P. duclauxi* were forms isolated from dairy products in contrast to the other species which grew less characteristically. *P. italicum*, and *P. luteum* with most of its allies showed little if any ability to utilize the lactic acid.

*Lactose.* Thom used lactose as 3 per cent of the mixture. It produced normal and quickly growing colonies of *P. camemberti*, *P. caseicolum*, *P. granulatum*; delayed but recognizable colonies of *P. chrysogenum*, *P. brevicaulis*, *P. citrinum*, *P. expansum*, *P. duclauxi*, *P. pinophilum*; others grew but slightly and as unrecognizable growths.

*Levulose.* Thom used levulose as 2.5 per cent of the mixture. The tubes were inoculated with conidia of fifty-two strains of *Penicillium* and incubated for twenty-one days; thirty-five organisms produced typical

colonies with the reaction of the liquid alkaline to litmus. Nine showed fair growth but a persistently acid reaction; the remainder were poorly developed. The normal colonies included most of the common species of the genus.

According to Brannon (1923) neither glucose nor fructose is a superior tissue former although *Penicillia* utilize fructose somewhat more favorably than glucose.

*Lignin.* Cultures (Thom) of a series of species upon Czapek's solution in which lignin was offered as a sole source of carbon mostly showed feeble growth although a few colonies produced abundant conidia. While some of the lignin must have been utilized the amount was not appreciable when subject to chemical analysis (unpublished work of Dr. Max Phillips).

*Malic acid.* Malic acid added as 1 per cent solution was tested (by Thom) against spores of thirty strains, all of which germinated as seen with the hand-lens, none reached the usual colony; in three weeks a few produced about half the normal or usual vigor of colony compared to that on Czapek with 3 per cent sucrose; among these in order of greatest vigor were: *P. chrysogenum*, *P. frequentans* (var.), *P. stoloniferum*, *P. commune*, *P. expansum*, *P. spinulosum*, *P. camemberti*, *P. biforme*.

*Paraffin.* Tauson (1929) reported a *Penicillium* which developed on paraffin of a melting point range of 45° to 56°C. which was capable of utilizing 80 per cent of the hydrocarbon.

*Pentose.* In a study of the action of some selected fungi on 2 per cent pentose solutions, Peterson, Fred and Schmidt, *P. camemberti* and *P. roqueforti* had only slight action on arabinose and xylose while *P. glaucum* was very active, completely utilizing 4 per cent solutions of these sugars in from four to five days. Carbon-dioxide and mycelium represented the major portion of the sugars used.

*Quinic acid.* Butkewitsch (1923 and 1924) in a series of papers reported that the carbon of quinic acid was utilized by "*Citromyces glaber*." He also found that catechol, while toxic in moderate concentrations, may be completely utilized by *Citromyces*, when in low concentrations.

*Saponins.* Solacoin found that *P. glaucum* utilized the saponins as a source of carbon but sporulation was retarded. Reducing sugars are not found in the substrata.

*Succinic acid.* Succinic acid in 1 per cent solution was tested against 41 strains (by Thom) among which no typical colonies were produced but seven strains produced growth which indicated appreciable power to utilize the acid alone for growth.

*Sulfur.* In an investigation of the sulfur nutrition of *P. glaucum*, Armstrong used sulfates, thiosulfates, sulfides, sulfites, thiocyanates and persulfates. Potassium persulfate did not permit the growth of these organisms. The sulfates served as more suitable sources for sulfur while the thiocyanates were the least satisfactory. Hydrogen sulfide was produced in all cases except when sulfates were used. Sulfates appeared in the culture solutions as the end product of the action of the fungus on thiosulfates. Molecular sulphur was frequently found and occasionally tetrathionates as well as globules of sulfur were identified in the hyphae.

Guittonnen (1927) reported that *P. glaucum* was capable of oxidizing precipitated sulfur to hyposulfites and sulfates.

*Tannin compounds.* Van Beyma reports that *P. phaeoanthinellum* Biourge is an active destroyer of tannin compounds.

*Tartaric acid.* In cultures of *P. puberulum*, upon Raulin's solution, the tartaric acid of the medium soon disappeared (Alsberg and Black).

#### METABOLISM: PRODUCTS OF PENICILLIUM ACTIVITY

The following paragraphs summarize current information as to biochemical products and activities reported as products of species of *Penicillium*.

##### *Acid production*

The formation of acids as a result of the decomposition of organic substances has become a commonly recognized characteristic of many species of *Penicillium*. The earlier workers added litmus, phenolphthalein and other indicators to the media before inoculation. At a later date the culture solutions were tested with the H-ion indicators of Clark and Lubs and more recently by electrometric methods. The literature contains numerous references to named species as "fermenting" particular sugars and not other sugars. Such records represent qualitative tests and occasionally titration of the substratum in cultures grown upon various types of "synthetic" fluid to which particular substances were added as the sole source of carbon. Most of these reports, where at all quantitative, lump all acid production as total acidity. Special workers however have studied the formation of particular acids, by certain species of the group, and some have sought to put such acid production upon an industrial basis.

Oxalic and citric acids have often been isolated from carbohydrate cultures of *Penicillia*. Since 1922 gluconic acid has been found by a

number of investigators in cultures of *Penicillia* on solutions containing dextrose, sucrose and maltose as sources of carbon. The results of experiments carried out in the past six years by a number of workers indicate that where the carbon source is suitable, gluconic acid is as widely distributed a product of the oxidative changes of sugars induced by fungi as is citric or oxalic acid. The condition of the culture medium seems to be one of the factors which determines to a large extent the preponderance in the end products of each of these acids. Recent work points to the conclusion that the relation between these three acids in mold fermentation of carbohydrates is not as close as was once thought. Gluconic acid cannot be an intermediate product responsible for the formation of citric acid; oxalic acid is known to be formed through reactions in which citric acid does not appear at any stage in the process. Experience has established that generalizations regarding acid production by fungi, even within a definite group, can be formulated with any degree of accuracy, only after careful experimentation with many strains, and in cultures where conditions are varied widely. Even with what is believed to be a carefully selected strain, the biochemical activity is apt to show great quantitative variations in successive mass transfers, and it is probably unwise to take anything for granted in this respect. Unpublished work in connection with the formation of gluconic acid by one *Penicillium*, (no. 2670), shows that variations in yield of from 10 to 65 per cent of the theoretical, occur with apparently identical cultures and under exactly similar experimental conditions. In the industrial application of any of these acid fermentations, it becomes of utmost importance to select carefully, strains showing vigor, both from the standpoint of growth and of biochemical activity. Constant attention must be given to culture conditions and to rechecking, if these characteristics are to be perpetuated with such a degree of reliability as to maintain an efficient industrial process.

The first of these studies of acid production of *Penicillia* goes back to Wehmer who described his two strains as species of a new genus *Citromyces* in 1893 and attempted to place the production of citric acid by them on an industrial basis. Many difficulties were encountered among which were length of time required and the necessity of carrying out the fermentation in the presence of calcium carbonate. The process has apparently never been utilized to any extent. Wehmer later reported a variety of *P. luteum* which produced appreciable quantities of citric acid from sucrose (*Chem. Ztg.*, 21, 1022, 1897).

Filosofov and Molinovski (*Nauch. Zapiski*, 5, 235, 1928) have studied

the ability of three types of *Penicillium* together with *A. niger* and *A. clavatus* to produce citric acid from sucrose, using Henneberg's solution with 10 per cent sugar. In twenty days their *Citromyces* (a monoverticillate *Penicillium*) produced citric acid equal to 17.1 per cent of the sugar in the medium.

Martin in 1916 reported a process for producing citric acid by the activity of our no. 28. The method does not appear to have become commercially successful. Our stock under that number was lost and never recovered.

*Citric acid.* Buchner and Wüstenfeld used a bean decoction diluted to contain 0.2 per cent nitrogen, to which was added calcium carbonate and 10 to 15 per cent of glucose. *Citromyces citricus* in this medium gave a yield of over 50 per cent in fourteen days.

Currie, about 1914, in collaboration with us surveyed the possibilities of commercial success with "*Citromyces*" and discarded the group as impractical in factory production of citric acid, substituting *A. niger*.

Butkewitsch (1923) noted that cultures of *Penicillium glaucum* on sugar solutions containing calcium carbonate and a low content of nitrogen produced in addition to citric acid an unidentified organic acid which formed a soluble calcium salt. This acid was later identified as gluconic acid.

The same investigator (1922, vol. 131) found that *Citromyces glaber* formed citric acid as a normal product from solutions in which an excess of all necessary nutrients were present.

Butkewitsch (1923) found that citric acid was not produced by *Citromyces glaber* in a medium containing quinic acid as a carbon source from which he concluded that citric acid is not formed by utilization of plasma substances but from the sugars themselves. In a further study of the metabolism of quinic acid by fungi he reported that *Citromyces glaber* when grown on salt solutions containing quinic acid, formed among other products 3-4 dihydroxy benzoic acid and catechol in the early stages of growth. These substances later disappeared, oxalic acid appearing in increased quantities (1924).

Butkewitsch (1925) investigated the conditions governing the formation of gluconic and citric acid from sucrose by *Citromyces glaber* and *Penicillium glaucum* and reported that low acidity favored the formation of the former and high acidity formation of the latter acid. More recently (Über säurebildung bei den Pilzen, in Bioch. Zeitschr., 182, 99, 1927) he has published the results of experiments with three different *Penicillia* and under the culture conditions described in his paper found both citric and gluconic acid present in varying quantities.

Chrzaszcz and Tiukow using a sweet beer wort as culture medium investigated the acid production by forty-six strains of *Penicillia*. They reported that while citric acid was definitely present in all the culture solutions of the species studied excepting three, the quantity of acid formed was chiefly dependent on the species. Oxalic acid formation was found to be irregular and this acid was recovered from the solutions in only small quantities. In some cultures considerable quantities of an acid were formed having a soluble calcium salt but the acid was not definitely identified.

In the course of a survey of the production of acid from glucose by fungi, May, Herrick, Thom and Church reported that many varieties of *Penicillia* formed appreciable quantities of various acids. Citric and gluconic acids in varying quantities were isolated from cultures of *P. divaricatum* and *P. citrinum* while seven different strains of the biverticillate series produced large quantities of gluconic acid only. One strain of the latter group (no. 2670, Thom) was active enough to warrant an investigation of the possibilities of its industrial utilization and a study was carried out to determine the conditions essential for a high production of gluconic acid. Herrick and May reported yields of 55 to 65 per cent of that theoretically obtainable from the glucose present in the culture medium when the oxidation was carried out under the following conditions: temperature 25°C., concentration of commercial glucose 20 to 25 per cent, nutrient elements in the following concentrations, N 0.016 per cent supplied as  $\text{NaNO}_3$ , P 0.00086 per cent as  $\text{Na}_2\text{HPO}_4$  or  $\text{H}_3\text{PO}_4$ , K 0.0026 per cent as KCl and Mg. 0.0024 per cent as  $\text{MgSO}_4$ . Citric or oxalic acids were never found in appreciable quantities.

Nierenstein, (1915) identified ellagic acid among the end products formed by a species of *Penicillium* which was cultured for twenty-two days on a 2 per cent solution of gallyl glycine.

*Mycophenolic acid.* Alsberg and Black using *P. stoloniferum* Thom (Thom and Church, no. 27) isolated and described mycophenolic acid suggesting the formula  $\text{C}_{17}\text{H}_{20}\text{O}_6$ ; this "behaves like a weak dibasic acid" and is non-toxic.

*Oxalic acid.* Many species of *Penicillium* produce oxalic acid as an end product of metabolism. Alsberg and Black report "minute" amounts as produced by *P. puberulum*.

Currie and Thom described *P. oxalicum* as producing oxalic acid in excess of other organisms tried. When calcium carbonate was added to the media the mold grew poorly but the yield of acid was greatly increased reaching at times 40 per cent of the sugar employed. Oxalic

acid so produced is not an end product but reaches a maximum in eight to twelve days and then diminishes. *P. oxalicum* produces acid from sucrose, lactose, potato starch and peptone.

Butkewitsch (1922, vol. 129) studied various salts of organic acids as carbon sources for the species of *Citromyces* and found the development of the fungus and the formation of oxalic acid was best with sodium salts rather than ammonium salts of the acids. He found that sodium salts of quinic acid were an especially good carbon source for oxalic acid formation in the case of *P. glaucum*.

*Penicillic acid.* Alsberg and Black using *P. puberulum* (see no. 157) found it to produce penicillic acid for which they suggested the formula  $C_8H_{10}O_4$ . It behaved like a monobasic acid; "it was toxic to animals when injected in a dosage of about 0.2 to 0.3 gram per kilo of body weight." The acid was most abundant in cultures grown under reduced air supply and in an acid substratum.

The work of these various groups has covered only a small fraction of the field of acid production by *Penicillia* and pointed to future possibilities in many directions with great academic interest and many industrial possibilities.

#### ALCOHOL SERIES

*Ethyl alcohol.* Alcohol is reported as produced by many species of *Penicillium* but never in more than very small amounts. Alsberg and Black found alcohol in growing *P. puberulum* upon sugar containing media. *P. glaucum* as a producer of alcohol was reported by Diakonow in 1886. The work of Kostychev on the production of ethyl alcohol by *P. glaucum* is mentioned in the preceding chapter.

Mannitol was found by Alsberg and Black among the metabolic products of *P. stoloniferum*. Bourquelot reported finding 2.5 grams of mannitol per kilo of mycelium in *P. duclauxi*.

#### COLOR PRODUCTION

The biochemistry of color production has been studied in a few species. Brenner reported upon work with *P. purpurogenum*. Canilov studied *Isaria virescens* which presents a nearly related problem, Martini and Dérivé-Degardes studied a species related to but not believed identical with *P. herquei*. Meyer studied the color produced by *P. variable* Wehmer as a substance coloring the cells yellow but not secreted as crystalline matter. Its production was closely related to the composition of the substratum. Sartory discussed the color production of



several species of "Citromyces" in which he limited his data to reaction and solubility in routine reagents.

Barber (1927) has reported the formation of a deep crimson color by an unnamed species of *Penicillium* when cultured on sucrose solution. The color was extracted with alcohol and was soluble in dilute sodium carbonate solution from which it is not extractable by ether. It gave an insoluble lead compound and was precipitated by acids after which it was soluble in ether.

Color production in cultures as factor in the identification and description of species is discussed in Chapter VI.

#### PRODUCTION OF ENZYMES

The *Penicillia* have not been so widely or so thoroughly investigated as to their enzymic activities as the *Aspergilli*. The literature shows that most of the work has been done with more or less ill described organisms designated *P. glaucum* which renders the value of much of the work questionable.

Cosmopolitan distribution and omnivorous habit are indications of the production of a multiplicity of enzymes. Many of the species of *Penicillium* are represented by closely related strains in many lands and found constantly upon the most diverse substrata. Few of them have been carefully studied but those that have been investigated have shown large enzymic capabilities. Whole series of molds, are presumed to produce invertase, various proteases; amylase, maltase, catalase, oxydases, etc. Miscellaneous references to activities of this kind are widely scattered in the literature but accurate study of known species by standard methods are much less common.

*Amylase.* Karrer (1921) has reported that the amylase of *P. italicum* gave maximum activity from pH 3.0 to 4.5 and was inactivated at pH 8.

*Cellulose destruction.* McBeth and Scales tested a series of species of *Penicillium* for cellulose destroying power using cotton and precipitated cellulose and reported the activity as present in *P. claviforme*, *P. luteum*, *P. stoloniferum*, *P. rugulosum*, *P. africanum*, *P. pinophilum* and "*P. roseum*." When they tested *P. africanum* and *P. pinophilum* upon rye straw and cherry wood shavings the cell walls were not attacked.

Scales (Bot. Gaz., 1916) tested thirty-one species from our collection for cellulose destroying power in petri dishes of cellulose agar. An enzymic zone of cellulose destruction was produced by twenty-three of these strains and absent in cultures of seven of them. Five of the strains

failing to show cellulose destruction in this experiment were originally isolated from Camembert cheese and are most commonly encountered in the dairy work. The other two, however, *P. lilacinum* and *P. divaricatum* represent series commonly found in the soil.

**Inulase.** Pringsheim and Perewosky (1926) studying inulase formation by "*P. glaucum*" found the mycelium to have more inulin splitting power when the mold was grown upon sucrose solutions than when the colony was grown upon inulin. Inulase production was proportionate to the vigor of colony development rather than a response to the stimulation of the presence of the inulin. Inulase continued to increase up to the 22nd day then decreased. Declerck (Bull. Assoc. école Sup. Brasserie Louvain, 25, 160, 1925) has found *P. griseo-roseum* hydrolyzes and utilizes inulin.

**Rennin.** Coagulation of milk is frequently reported (Sopp, Thom and others), but without analysis of the observation to separate acid curdling from rennin curdling or the activity of some other protein splitting enzyme.

**Saccharase.** According to Kertesz (1928), *P. glaucum* contains saccharase only when grown on media containing carbohydrate in which the alpha fructoside structure occurs.

Waksman and Davison (Enzymes. Williams & Wilkins, Baltimore, 1926) review the more reliable literature up to 1924 and the following table has been abstracted for the most part from their work.

Amidase.....	<i>P. camemberti</i>	
	<i>P. pinophilum</i> .....	Clark and Scales
Amylase.....	<i>P. duclauxi</i> .....	Bourquelot and Graziani
	<i>P. glaucum</i>	
	<i>P. pinophilum</i> .....	Clark and Scales
Cellobiase.....	<i>P. purpurogenum</i>	
Cytase.....	<i>P. atramentosum</i> .....	Sanborn
	<i>P. chrysogenum</i> .....	Sanborn
	<i>P. expansum</i> .....	Sanborn
Diastase.....	<i>P. camemberti</i> .....	Dox, Robbins
Emulsin.....	<i>P. camemberti</i> .....	Dox
	<i>P. glaucum</i>	
	<i>P. pinophilum</i> .....	Clark and Scales
"Glykose oxydase".....	<i>P. glaucum</i> .....	Muller, 1928
Invertase.....	<i>P. glaucum</i> .....	V. Euler, Kopeloff and Byall
Inulase.....	<i>P. camemberti</i> .....	Dox
	<i>P. glaucum</i> .....	Pringsheim and Perewosky
	<i>P. griseo-roseum</i> .....	Declerck
Lactase.....	<i>P. camemberti</i> .....	D

Lipase.....	<i>P. camemberti</i>
	<i>P. glaucum</i>
	<i>P. pinophilum</i> .....Clark and Scales
	<i>P. roqueforti</i>
Maltase.....	<i>P. camemberti</i> .....Dox
	<i>P. glaucum</i>

*Fat production.* Barber working with an unnamed species of *Penicillium* in sucrose solutions reported a light yellow fat in considerable quantities. In our own cultures the certain strains with the aspect of Corda's *Clonostachys araucaria* (see no. 450) produced masses of mycelium apparently full of fat globules.

*Mycodextran.* Dox and Neidig in 1914 reported and described mycodextran as a new polysaccharide produced by *P. expansum*.

Gagerov found that *P. crustaceum* caused a marked cleavage of phosphorus pentoxide from phytin.

*Starch production.* Boas (1917) reported a starch-like substance as produced in sugar solutions containing ammonium salts by *A. niger* and *Penicillium* sp.

Chrzaszcz and Tiukow investigated the formation of starch from sucrose by 45 species of *Penicillia* and concluded that the formation of starch-like substances by these organisms was a wholly normal process, the starch serving as a reserve food stuff. They grouped these *Penicillia* into two classes, the starch formers and the acid formers and hold that the former are older evolution forms than the latter.

#### SULPHURIC ACID

Cappuyns (1926) reported that certain molds including species of *Penicillium* when grown upon a medium containing ammonium sulphate break up the salt, use the ammonia and set free sulphuric acid. In his experiments the resulting concentration of  $H_2SO_4$  reached 4 grams per liter. Wehmer reported the same general observation in 1913.

#### ARE TOXINS PRODUCED?

Zipfel (1894) fed dogs, hens and goats with feeding stuffs in which "*P. glaucum*" had been allowed to develop until the products were "moldy." No evidence of toxicity was obtained. Similar studies of moldy feed by Church and Buckley were also negative. Turesson however reported toxic effects from feeding *P. avellaneum* and *Paecilomyces* (*P. divaricatum*) to rabbits.

## TOXICITY TO PLANTS

Behrens studying the rotting of fruit (1898) reported that toxic substances are produced which weaken or kill the cells of the host plant.

Sturli (1908) described a toxic substance from "*P. glaucum*."

Barnum (1924) found that *P. expansum* when grown in various modifications of Czapek's solution produces a wilt inducing substance which is not destroyed by boiling, autoclaving, addition of acid or alkali or dilution.

## UREA

Fosse detected urea in the cell contents of *P. glaucum*. According to Ivanov, (Excretion of Urea by Fungi. Bioch. Zeitsch., 157, 229, 1925) the formation of urea by fungi, including *P. glaucum*, is dependent on the composition of the culture medium. Peptone and gelatin serve as sources for this compound. The urea appears to be a product of excretion and diffuses into the medium. Urease may or may not appear depending on the carbon and nitrogen relations in the nutrients.

## CHAPTER IX

### OCCURRENCE AND SIGNIFICANCE IN NATURE AND INDUSTRY

The physiological factors involved in the appearance of the penicillate molds under natural conditions or in connection with processes of industrial importance have already been considered (Chapter VII). References to "Penicillium sp.," "some" Penicillium," "*P. glaucum*" or to some other species clearly inadequately identified are common in publications discussing the fermentation industries, the production, preservation, and distribution of food, feeding stuffs, of fibers and textiles, of seeds, bulbs and of miscellaneous substances.

The following pages will present summaries of current information with special reference to those phases of Penicillium activity which have been actually handled by us in the laboratory. Topics are grouped and arranged alphabetically to group.

#### BOTTLE IMPS

When liquid media are inoculated with many species of Penicillium and stored for considerable periods great differences are observed between the resulting colonies. Some species produce a simple mycelium covering the surface and bearing a single crop of conidia, then die or at least cease growing. The mycelia of other species continue to grow, buckling, twisting and forming fantastically shaped masses which often follow the drying of the fluid to the bottom of the vessel. In closed containers infected and let stand for long periods these mycelial masses sometimes assume remarkable forms. In one case in a bottle of lime-juice (no. 4172) the mycelium formed a tubular mass growing apparently at the top and pushing toward the bottom of the bottle several inches below. This organism was an unidentified monoverticillate Penicillium. Such growths (often called "bottle imps") are occasionally encountered in fruit juices and soft drinks of various kinds. Attempts to produce the peculiar tubular forms by inoculation have failed although fantastic growths are not uncommon in old cultures upon liquid substrata and a short tubular form was found in one of our experiments. Similar observations are reported by Bachmann.

## BULBS

Rotting of the bulbs of lilies, hyacinths and related families, in storage, has been a serious loss to florists and gardeners. In the course of the inspection of imported bulbs by the Federal Horticultural Board, species of *Penicillium* have been reported to us for several years. Recent studies of such importations extended also to domestic bulbs have shown (McCulloch and Thom) that a particular sclerotium producing *Penicillium* (*P. gladioli*) is often associated with rot of gladiolus. Strains of this species were independently isolated by the describers in Washington, by Mr. O. H. Elmer, in Manhattan, Kansas, and by J. E. Machacek in Canada. Miss C. A. Pratt in England working with hyacinth bulbs isolated and sent us two strains (no. 4811) one of which seemed to reproduce Biourge's description of *P. hirsutum* Dierckx the other shaded from this species toward *P. cyclopium* Westling. Comparative work with a series of bulbs subsequently confirmed the conclusion that *P. cyclopium*, *P. palitans* and *P. hirsutum* belong in nearly related series in the fasciculate group in which zonation is absent or only slightly developed. Numerous studies over a period of years had already yielded cultures of *Penicillium* as associated with rotting bulbs. Comparative study has brought recognition that the *P. cyclopium* and *P. hirsutum* series of forms are active agents in this destruction.

## CABBAGE BUDS

Two strains of *Penicillium*, one rather suggesting *P. ventuosum* Westling and the other *P. stoloniferum* were isolated by Edwin E. Honey in 1922 from cabbage buds.

## CEREALS AND CEREAL PRODUCTS

Cereal grains and their products present highly starchy substances containing a certain amount of sugars and ample stores of protein and salts, hence are exceedingly favorable substrata for *Penicillia*. Cultures of such products over many years show the abundant presence of spores or mycelia of species of *Penicillium* along with *Mucors*, *Aspergilli*, *Cladosporia* and many other molds (see Thom and Hunter, Thom and LeFevre and numerous authors). One needs but to add water to a little above the critical percentage for stability to obtain abundant growth of such organisms.

References to such growth as mold, or as vague references to members of this genus among other genera are numerous in the literature of the

grain, milling, and baking industries. A few only of these discussions may be cited here as they apply to particular products.

### *Flour and meal*

Mill products in general are not sterile. Cultures usually show a wide variety of bacteria and molds including species of *Penicillium* (Thom and LeFevre, Frazer, Prescott et al.). In comparison with the distribution of the organisms in the various fractions, the highest numbers of colonies found in culture appear in the fractions containing the outer layers of the grain such as bran and from the germ; the numbers in the finely ground portions (flour of various grains) follow apparently as a result of aspirating systems which carry the finest dust into such products. The lowest numbers are in coarser masses (hominy, grits, etc.) consisting of uncrushed masses of the horny endosperm. The more finely ground products have but to acquire a slight amount of extra moisture to develop lumps or mold balls with attendant musty odors.

The mill, the bakery, the hands and the clothes of the work men are usually well dusted with the cereals in use. The products manufactured, such as baked or packaged goods, even though supposed to be sterilized, usually carry mold spores into the lines of distribution. Such products require only favorable conditions of growth to develop mold colonies.

Mustiness involves such losses that many investigators have sought means for prevention and for the renovation of the damaged materials. Ludwigs (1926) offered formulas for the prevention of mustiness in cereals which he regarded as resulting from the activities of *Aspergillus glaucus*, *Penicillium glaucum* and similar species.

### *Bakery products*

In general the baking process carries the dough of bread, cake, pies, etc., well beyond the thermal death point of *Penicillia* (Thom and Ayers). Nevertheless losses are frequently encountered.

*Bread.* *Penicillium* grows readily on bread, competing with *Aspergillus* and *Mucor* as a cause of losses. Properly baked bread is free from *Penicillium* either in vegetative or conidial form as it comes from the oven (Prescott et al.). *Penicillia* as a visible factor in moldiness of bread only appears as a result of infection after baking commonly in cooling and in handling in connection with distribution. Frequently handling involves wetting of the surface with condensation water while the bread is cooling. This is followed by holding for considerable time in con-

tainers which are often poorly ventilated. *Penicillium* upon such loaves appears as superficial green patches.

Only a few species seem able to develop in the interior of a loaf. A loaf wrapped in sterile paper or dipped in hot paraffin as it comes from the oven may be held free from mold. In preparing bread (Thom, 1906) for use in inoculating Roquefort cheese such loaves are inoculated by thrusting a pipette with a suspension of spores into the center and depending upon tolerance of this species for low tensions of oxygen to bring about its development and sporulation throughout the loaf. In many experiments we have maintained the culture pure by this means.

### *Corn*

Many studies of molds as a cause of deterioration in corn (maize) have shown members of the *Penicillium* group as active agents in loss. Our own studies have shown, however, that such loss is directly connected with the presence of moisture in percentages above that required for proper storage. In our experiments the *Penicillia* were found unable to grow at percentages of moisture at which members of the *Aspergillus glaucus* group were still active, hence the latter series becomes the test series to determine the moisture content for safe storage. In practice, however, several *Penicillia* are encountered in spoiled or spoiling corn. *P. oxalicum* has been isolated frequently, a yellow *Penicillium* was found present in all the grains of a sample once received from Ohio; monovericillate types are frequently found in infected grains of corn.

### *Wheat*

Hurd studying the susceptibility of grain to mold injury reports that the unbroken seed coats of wheat or barley prevents the entrance of *Penicillium* in damp storage, in soil or in blotted germination, but if the seeds are cracked in locations which expose the endosperm, infection takes place readily.

Defective germination of wheat due to the invasion by *P. glaucum* through cracks in the seed coat made by harvesting and threshing operations was reported from New South Wales by Noble. He found that treatment with copper carbonate dust designed to control smut also controlled this form of damage.

### CHEESE RIPENING

The Camembert-Brie group of cheeses originating in northern France obtain their texture from the proteolytic activity of *P. camemberti*



Thom (Dox, Thom, Mazé). These cheeses are made in thin cakes, perhaps  $1\frac{1}{4}$  to  $1\frac{1}{2}$  inches (3 to 4 cm.) in thickness and varying in diameter. The freshly made curd is salted upon the surface, inoculated with the mold or placed in a room in which the spores of the mold reach the surface from their abundance in the atmosphere. The cheese contains 55 to 60 per cent moisture at the outset and is exposed upon mats or boards in a humid ripening room at temperatures preferably from 50° to 60°F. (optimum at 13° to 14°C.), in which the entire surface becomes covered in about a week with a floccose white covering of mold, perhaps 1 to 2 mm. deep. In about ten days the mold shows a bluish or greenish gray from developing conidia. After about ten days, the curd in contact with the mycelium begins to soften to a smooth buttery texture which gradually extends from all sides toward the center of the mass. The curd at first markedly acid to litmus becomes alkaline to litmus as the softening progresses inward. In a period varying with the water content of the mass, the temperature and humidity, but commercially about four weeks, the entire mass is softened. In Dox's studies the change was found associated with a proteolytic enzym similar to erepsin which produced abundant caseoses but no putrefactive products (see the taxonomic section, Chapter XVI for the synonymy of the mold and Chapter VIII for its enzymic activities).

A nearly related species, *P. caseicolum* Bainier, promoted by Roger for ripening the same types of cheese lacks the green color in its conidial areas (see synonymy in taxonomic section no. 202). This species develops in the same manner as *P. camemberti*, but produces a cheese of somewhat different texture and less attractive flavor. The physiological adaptation of these two molds to the cheese industry is so close that they are thus far unknown outside the dairy. They grow readily in culture. They may be propagated in mass on crackers for inoculation of cheese, or the starting of new factories. In the factory, they dominate, overgrow and suppress competing forms if the cheese falls within a fairly narrow range of water content, the humidity is maintained around 88 per cent, and the temperature about 50° to 60°F. Any wide departure in either particular is commonly followed by the development of other species to the detriment of the resulting cheese.

Such green forms as *P. commune*, *P. roqueforti*, *P. biforme* give a bitter taste and a musty odor. Varieties of the *P. brevicaulis* group of organisms are usually present to some extent and under conditions unfavorable to the Camembert mold, overgrow it and injure the product. The presence of *P. brevicaulis* in the cheese room is accompanied by a strong am-

moniacal odor. Ammonia production from such growth is often strong enough to turn a moist litmus paper held above, but not in contact with the cheese. *P. brevicaulis* when abundant on cheese gives a strongly ammoniacal taste to the product.

### *Roquefort*

In contrast to the Camembert mold, *P. roqueforti* is a velvety green species which invades the holes and cracks in loose textured cheeses. In manufacturing such cheeses as Roquefort, Gorgonzola and Stilton, the curd is so managed as to leave cracks, channels and openings between the particles as they are pressed together. In some varieties additional holes are punched to let air penetrate throughout the mass. Such cheeses when ripe show "marbling" with green mold on the cut faces. In studying these cheeses, Thom and Currie showed that these channels and spaces within the cheese contained less oxygen than atmospheric air and that the Roquefort mold obtained its ability to dominate the flora of these spaces from its ability to grow with oxygen composing as little as 5 per cent of the gas mixture present, whereas in their experiments covering twenty-one species of *Penicillium* and five of *Aspergilli*, *P. roqueforti* alone could tolerate the low-oxygen tension demonstrated to occur in the cheese. Currie further showed that the characteristic flavor of this group of cheeses was due to the splitting of the butter fat by this organism to produce a different mixture of fatty acids from that produced by bacterial activity.

The molds found in Roquefort cheese made from sheep's milk in southern France, in Gorgonzola cheese made in Lombardy, and in Stilton cheese made in England belong in a single series, although Sopp, Biourge, Arnaudi, and others have attempted to draw up descriptions which will make them into separate species. Steuart and later, Golding working with the whole group of "blue-veined" English cheeses has found the selection of particular varieties or strains of mold and consistent inoculation with them desirable since under the working conditions of the English cheese factory undesirable varieties or species do complicate production. The successful utilization of such a species thus depends upon close control of the conditions of growth. Such control has only been developed in handling a small number of varieties of cheese. In other varieties, mold ripening is uncertain and unreliable.

### *Cheese rind*

Staub (1911) observed and described *P. casei* as the cause of black spots on and just under the rind of Emmentaler cheese in Switzerland.

This was conspicuous and was isolated by Thom from Swiss cheese imported to America about that time but efforts to obtain it in America have not been successful in recent years.

#### COAL AND COAL RESIDUES

Fisher and Fuchs report *Penicillium* as appearing upon certain samples of German coal and upon residues and extractions from coal. The mold grew more readily in these experiments upon treated or extracted samples than upon raw coal direct from the mines. The species of *Penicillium* involved was not identified. The experiments did not go far enough to show exactly what components of the particular types of coal studied, were able to support mold growth and which were inhibitive.

Iwasaki reports *Citromyces pfefferianus* as growing on coal products such as coal powder and blocks, as growing more readily on dull coal than bright, as growing more readily on brown lignite than black lignite and as probably never growing on true coal. The growth of the fungus is never thick and luxuriant at the best.

#### COPRA

Molds are reported by Eaton and by Brill, Parker and Yates to grow on copra having a moisture content as low as 5 to 7 per cent. The chemist's figure as far as the growth of *Penicillia* is concerned is only a crude average of the moisture in the center and the surface of the sample. Common molds are supported by superficial areas and pockets of moisture. *Penicillium* in the experiments of Brill and his colleagues did little damage.

#### COCOA

Contaminations of fermenting vanilla beans may come from the vats, the bottoms and walls of which furnish different species of *Penicillium* (Ciferri, 1927). A collection of such molds from the fermenting masses of beans in the Gold Coast Province of Africa was submitted by Mr. L. J. Schwartz of the Bureau of Chemistry while studying the handling of this product. There did not seem to be any specific *Penicillium* active in such spoilage. They were present rather as a result of careless handling.

#### CORKS

The occurrence of serious losses in bottled grape juice directed attention to the contamination of corks and compounds of cork used in lining

bottle caps. These were commonly found to carry mold spores if not actual growing colonies. Some of these stoppers could be sterilized; others by their nature would be ruined by either steam or dry heat. Such stoppers dampened with nutrient during filling and handling and allowed to stand upon the shelf became covered with mold producing spores which dropped upon the surface of the contents of the bottle and developed extensive colonies. Losses of this kind were avoided by treatment of the cork with innocuous substances such as paraffin which would not support mold growth.

#### EGGS

Molds including *Penicillia* have been reported frequently from eggs. Some of the earlier papers merely listed organisms found. Others attributed specific importance to every organism found (See Artault, 1893; Gueguen, 1898). Weston and Halnan noted that as long ago as 1864 it was shown by Mosler that uninjured eggs may be infected from the outside with "*Penicillium glaucum*." In their own experiments on the conditions necessary for penetration they state that damp nesting boxes containing straw is all that is required for contamination of eggs with *Penicillium* sp. In other words, *Penicillium* spores on the surface of the dry clean egg are unable to penetrate the shell, but hyphae can gain easy entrance through pores of wet shells.

Species of *Penicillium* have been occasionally encountered in our own examination of moldy eggs but without finding a particular species in anyway consistently associated with the damage, which is rather one of bad handling than specific infection.

#### *Egg shells*

*Penicillia* have also been reported as forming colonies discoloring museum collections of shells. In these cases, high humidity had brought the organic constituents of the shell and its membrane to a moisture content capable of supporting such mold growth.

#### ENSILAGE

*P. roqueforti* and related strains are the characteristic molds of silage when exposed to the air. A surface layer to the approximated depth of about six inches becomes completely overgrown with fruiting colonies within a few days. The same species is commonly found in holes and pockets wherever air has crept in on account of careless packing in the silo. Other species may be isolated when dilution cultures are made but

in many studies made in the silo and in the laboratory, the predominance of *P. roqueforti* has been quite general. Reed and Barber report *P. italicum* as also present but we have never found it as a significant species outside the citrus industry.

#### FAT DECOMPOSITION

Penicillia appear as causes of deterioration in fat only if water and other nutrients are present in varying quantities in the mass. Animal fats occasionally show spots of mold but careful examinations prove that tiny globules of water charged with other nutrients were distributed within the mass. Laxa reported Penicillium as breaking down butter-fat. Mold in butter (Thom and Shaw) is conspicuous and often causes considerable losses. Butter, however, contains 14 to 16 per cent of water (brine) with considerable amounts of milk proteins, milk sugar and the salts of the milk serum. In salted butter the concentration is commonly great enough to restrict the mold flora. Enclosure of the watery components in fine globules and films within much larger masses of fat restrict greatly the number of species capable of growing. *P. roqueforti* is commonly present. The same species has been found by us in oleomargarine along with Paecilomyces (*P. divaricatum* Thom). Other discussion of such activity has been found in Crampton, Rahn, Thom, Seliber and Zikes' papers.

When other nutrients were furnished (in Czapek's fluid) Thom grew various species on purified butter fat as a sole source of carbon. This fat was attacked readily enough by various species to permit the development of normal mold colonies.

#### FIBERS

##### *Abaca*

Serrano reports that complaints of mold deteriorations in abaca (Manila hemp, *Musa textilis*) from the Philippines had been received since 1920. In his studies fungi were isolated from the deteriorated abaca and inoculated back into sterile pieces of fiber. After about two months of incubation it was noted that the organisms causing the greatest amount of deterioration were of the cellulose destroying type among which was "*Penicillium glaucum*." A histological comparison of sound and unsound fibers showed in the latter a disorganization of tissue not observed in sound fibers, fungous infection being 80 per cent as compared with 5 per cent of the former. Serrano recommends that all fiber be

dried to a moisture content of 11 per cent or 12 per cent before baling and storing. Warehouses for storing abaca present conditions decidedly favorable to the development of fungi. Therefore Serrano recommends that these be more adequately ventilated and care taken in handling.

### *Cotton*

Penicillia are reported by various students of mildew as a cause of damage to cotton fabrics. Various species are easily found in examining mildewed cloth but when put back upon experimental samples the species tested have usually proved unimportant as causes of the weakening and discoloration associated with the name mildew. The whole subject with its literature is discussed by Armstead and Harland, and by Bright, Morris, and Summers, but without measuring specifically the damage attributable to the Penicillia.

Burns in 1927 claims to have isolated *Penicillium africanum*, *P. roseum*, and *Penicillium sp.* from seed cotton which had been stored under slightly damp conditions for three months and which had been exposed to a slight rainfall three days prior to being sampled. He continues stating that in his belief colored fibers due to fungous growth reveal themselves "in patches in the bale particularly to the depth of 3 or 4 inches." Comparatively high numbers of fungi were found in damaged lint. Fungous infection as a source of serious deterioration of cotton during damp storage is to be sought and controlled in the unginned rather than in the ginned material. Extensive references are appended for deterioration and microscopy of cotton cellulose-decomposing bacteria and fungi, etc.

### *Flax*

Ruschman in a study of flax retting and the preservation of the fiber finds *Penicillium* (sp. ?) one of the molds connected with loss. He notes that the commercial standard of 12 per cent moisture in this product is too high to cut out mold growth.

### *Textiles*

Two samples of fabric used on airplane wings about 1921 showed on examination *Paecilomyces varioti*. One of these samples showed also a strain of a green *Penicillium* giving some rosy color in Czapek solution agar. Microscopic examination of the browned, mildewed spot of the latter sample showed mold hyphae among the fibers. No tests were made to show whether the fiber was weakened or not by the activity of these molds in the samples.

Morris working upon mildewing of cotton goods found *Penicillia* to be a common cause of mildew of sized goods. The sizing used is variously manufactured of tapioca flour, sago, farina, maize, and cassava. Such products are not uniformly attacked but in general present favorable media for mold growth with resultant spotting and ultimate weakening of the fabric.

Bright, Morris and Summers conclude that when once unpacked or stored in the warm and humid conditions prevailing in most tropical countries, no cotton goods can be regarded as immune from attack of mildews. These authors find *Penicillia* producing both acid and alkaline conditions when spotting dyed and printed goods, irrespective of the condition of the original fabric. Acidity is produced most rapidly in sized fabric because of the carbohydrate present but it may also be produced slowly from mold growth affecting the cellulose. Weakening of fibers is doubtless due to enzymic action and not to acidity produced, the latter having been overemphasized.

#### *Paper and paper pulp*

A serious spotting of white "print" paper in the Bureau of Engraving and Printing at Washington occurred in 1924. In piles moistened to prepare them for the press numerous red spots developed at various levels in the pile. These became as much as 1 cm. in diameter, hence involved many sheets. When cultures were made *P. purpurogenum* var. *rubri-sclerotium* Thom developed in pure culture.

In wood pulp prepared and held for paper making, various species of *Penicillium* develop among a great variety of other fungi. These conditions were studied by Lodewich in Northern New York and Güssow in Canada, and by the Forest Products group at Madison, Wisconsin (Kress, Humphrey, Richards and Staidl). In their summaries of injuries due to fungi, *Penicillia* seem to be significant primarily as discoloring organisms attacking soluble carbohydrates rather than destroying or weakening the fibers themselves. To reduce such damages, series of antiseptics have been developed which are more or less practically effective.

*Penicillia* in moist, dirty, or rotting paper are incidental to miscellaneous contamination rather than indicative of a special type of spoilage in such material.

Güssow reports *Penicillium* as spotting bank note paper, and finished copper and steel engravings.

## FRUITS

The acid fruits are primarily attacked by molds rather than bacteria. Rots in these products when stored involved huge losses hence extensive investigations have been conducted by Experiment Stations, the United States Department of Agriculture, various research bureaus in England, South Africa and upon the continent. Some of these investigations, product by product, will be discussed together with our own studies of these activities.

*Apples*

Penicillium rot is one of the largest factors in the destruction of stored apples. Exhaustive search for the forms present in such spoilage brings to the laboratory many species and varieties but scrutiny of the effects in mass supported by extensive experiments by ourselves and many others show that the members of the *P. expansum* series are by far the most destructive. One estimate by the Bureau of Markets gave "blue mold" as the cause of 80 to 95 per cent of the losses from rot in transit and in commercial storage. Inoculation experiments show the soft rot areas are quickly formed and rapidly extend to involve the whole fruit. The development of these rotten areas under the usual storage conditions is accompanied by the formation of great coremia by the members of this group. These coremia vary from short stocky aggregations of conidiophores with clustered penicilli at their tips to stalks up to 1 cm. long and perhaps 2 mm. in diameter producing large heads of conidia. The conditions and organisms present have been studied by many investigators; Wehmer (Beitr.) figured these coremiform structures as *P. glaucum* in apples and grapes; Eustace (Geneva) studied the species in culture at storage temperature; Sopp incidentally mentions them; Brooks and his co-workers have studied the organisms and conditions for their control for many years. Ciferri in Italy described *P. malivorum* in terms which leave no doubt as to its relationship to *P. expansum* whatever its minor differences. Fisher summarizing these studies concludes that *P. expansum* can not penetrate the uninjured surface of the apple but recognizes that the injury required for entrance is so small that the prevention of infection is difficult. Kidd and Beaumont surveyed the whole field of losses in the fruit arriving in London. They isolated and studied many strains of Penicillium as active agents, mostly perhaps as incidental contaminations. Putterill in South Africa isolated many of these forms in the storehouses of Cape Town.

The earlier workers identified the Penicillium rot of apples with *P.*



*glaucum*. With recognition that this name was too loosely used the name *P. expansum* as suggested by Thom has been substituted by most groups and it has been shown that there are a series of varieties or strains grading into each other, but together constituting an aggregate species which has already been discussed as a major cause of storage rot of fruits of the apple family, apples, pears, *Mespilus*, etc.

Careful studies by Brooks and his colleagues have shown various other species of *Penicillium* to be occasionally present. In comparative experiments apples have been inoculated with many species of *Penicillium*. Some species produce rot in varying degrees; some are inactive on the apple, but no *Penicillium* unrelated to *P. expansum* as a species-aggregate has proved to have commercial importance as a cause of such rot.

*P. expansum* produces extensive soft rot areas going clear to the core of the apple and produces its conidial masses on the rotting area under the usual storage conditions as *Coremia* or scattered fruiting bodies consisting of white stalks composed of large numbers of conidiophores massed together and spreading at the apex to produce a "head" composed (as seen under the microscope) of penicillate conidial masses. Conidia transferred to culture media produce the *P. expansum* type of growth with coremium production indicated by clustered or fascicled conidiophores, but frequently inconspicuous. The stalks and green heads produced by this species upon the apple were probably the basis for Link's species, *Coremium glaucum*, and for part, at least, of Corda's figures of his species of *Coremium*.

The mechanism of attack in the destruction of fruit by *Penicillium* and *Mucor* was studied by Nobecourt who attributed the break down of tissue to enzymes secreted by the molds. These enzymes were rendered inactive by heat to 60°C. for fifteen minutes. Their activities are reduced or greatly delayed by temperature near the freezing point. Juice expressed from the rotten fruit had a strong cytolytic and plasmolytic action. Much further work must be done to clear up the limitations of the rotting activities of these fungi.

One author has suggested that rot organisms reached the tissue of the apple only after entering the cut end of the stem and growing through its full length. We doubt the existence of a decay fungus which enters sound apples through the stem of an apple after its removal from the tree. We would not doubt the possibility of such organisms entering through the calyx and spreading throughout the interior of the apple from that direction.

### Berries

Moldy blackberries collected by Miss Leva B. Walker of Nebraska in 1918 presented *P. luteum* and *P. rugulosum*, a form related to *P. oxalicum*, a member of the *P. roqueforti* group, *P. stoloniferum*, and *P. divaricatum*. Moldy red raspberries from the same source presented members of the soil group and of the *P. roqueforti* group. Moldy loganberries presented strains of the soil group and of the *P. luteumpurpureogenum* group.

### Citrus fruits

The destruction of citrus fruits by particular species of *Penicillium* was first fully discussed by Wehmer (1893) although *P. digitatum* as a rotting organism upon oranges had been described and distributed in exsiccati by Saccardo in 1881. Wehmer disregarded *P. digitatum* and redescribed it as *P. olivaceum* from its olive-green color upon the oranges in contrast to the blue-green of *P. italicum*.

Wehmer's studies became the starting point of extensive investigations of the conditions surrounding the infection of citrus fruits by these species. Powell and his co-workers in the United States Department of Agriculture showed that these species while always present in citrus producing areas are wound parasites rather than capable of penetrating the unbroken surfaces of the fruits. They thus were able to reorganize the whole scheme of distribution, thus effecting great reductions in the losses due to rotting in storage and in transit. More recently it has been found possible to wash these fruits in a weak boric acid and borax solution (Shiver, Fulton and Bowman), which seems to be a selective antiseptic against such losses. The borax treatment is reported to be more effective against *P. italicum* than *P. digitatum*.

Fawcett working in California contributed a pure white form identical with *P. digitatum* (*P. digitatum* var. *Californicum*) in activity, but lacking the olive color. These species are so closely associated with citrus fruits that they are rarely encountered elsewhere. Evans, Thompson and Putterill working in South Africa have made similar studies of the agents and causes of loss of citrus fruits due to rotting, especially in the holds of vessels and in the terminal warehouses of Cape Town. Carne in 1925 discusses the blue mold on oranges in Western Australia. Tarby in 1924 made a similar report from Fiji. These reports are indicative of the world wide presence of these molds in the citrus industry.

Fawcett and Barger compared the rate of development of *P. digitatum* and *P. italicum* on oranges at different temperatures when inoculated

at both the stem and stylar ends. They report that injuries near the stem end were generally greater than those near the stylar end and that *P. digitatum* does not develop at such extremes from its optimum temperature as does *P. italicum*. The optimum was the same for both organisms.

In addition to these species which are quite specific to the citrus industry, Nakata in studying the molds commonly found about the citrus industry also contributed to us for identification, a strain of the *P. expansum* group, a strain similar to *P. erectum* Bainier and *P. islandicum* of Sopp. Fawcett in California sent in a strain of the *P. roseum* group as occasionally destructive to citrus fruits.

Horne, on the other hand reports *Penicillium expansum* as causing an apple green mold of feijoa (*Feijoa sellowiana* Berg) but did not find the blue and green molds of citrus naturally developing, or with inoculation.

#### *Lemon juice*

*Penicillium italicum* was found by E. M. Chace in 1920 growing as white mycelium on lemon juice under cold storage conditions. Such spoilage in lemon and lime juices and similar products does not involve any danger to human health but does involve a loss of flavor and the production of objectionable flavors.

#### *Grapes*

Wehmer (1893) noted the presence of *P. glaucum* on grapes. Our own studies over many years show *P. expansum* in its coremiform phase to be the usual type of *Penicillium* rot upon grapes in storage. Investigations involving similar observations have been made in several countries. Some of these may be cited.

Blue and green *Penicillia* on grapes in France are reported by Mathieu. This moldiness occurred during a rainy season at the end of the summer. The grapes in question had been punctured by insects. Not only was the flavor of the grape pulp affected by the mold but also that of the must made from the moldy grapes; acidity and tannin were reduced, bouquet and color altered, and gummy substances and oxidizing diastases being formed.

Aguilera reports from Almeria, Spain, that grapes already packed in barrels become spoiled from *Penicillium* "glaucum" which attacks the stalks and fruit, and nearly always produces the first signs of rot. The mycelium of the *Penicillia* pierce the fruit causing it to become dark,

the pulp becomes soft, the skin soon appears slightly brown, with round spots which spread and in their centers, become whitish and then greenish blue. The cork used for packing the grapes is probably the greatest source of infection.

#### LEATHER

Incidental cultures of *P. rugulosum* and some other members of the biverticillate group have been found but without evidence of important destructive effects.

#### MEAT

In a series of moldy samples of beef trimming we found strains of *Scopulariopsis* to be fairly common. The green forms so abundant upon plant remains are absent or only obtainable by culture methods which point to spore contamination rather than colony production.

In cold storage the green *Penicillia* are frequently found in cultures from moldy meats but Brooks is probably correct in calling them incidental contaminations but not serious agents of decomposition. On the other hand, we have frequently isolated members of the *P. brevicaulis* (*Scopulariopsis*) group from moldy meat upon which there is occasional evidence of significant injuries.

#### Ham

In one case very large quantities of ham developed a peculiar, musty odor without other evidence of decomposition. Spores of *P. brevicaulis* were found upon the skin surface of the hams examined but not in large numbers. Cultures, however, made from various levels throughout the flesh consistently showed *P. brevicaulis*. Mustiness in hams has been reported to us from two sources. In this shipment in possession of the War Department in 1919, about two-fifths of the individual hams developed an odor comparable only to certain over-ripe Camembert cheese badly infected with *P. brevicaulis* (*Scopulariopsis*). The odor and taste persisted when the ham was cooked rendering the lot unsaleable but apparently without ill effect when eaten. Cultures from various levels in the flesh showed *P. brevicaulis* clear to the bone.

Causal relationship between the musty odor and the mold was not proved but the correspondence between the conditions already known in cheese and the ham left the relationship presumptive.

## NUTS

Penicillia are often found in moldy nuts. On account of their apparent stability nuts of many kinds are frequently carelessly handled. Sometimes they are gathered in damp condition and stored wet. The shells are commonly more or less contaminated with soil with its load of bacteria and mold spores. As the nuts are gathered into piles or containers ideal conditions for mold growth are produced. Such organisms as Penicillia find ready entrance to the interior through cracks in the shell.

The amount of damage done as well as the agent capable of injuring the meat depends upon the condition of the nut-meats themselves. Low water content is essential in the prevention of damage. This water content must further be rather a percentage of water calculated against soluble or fermentable substances rather than an absolute percentage for many nuts are very high in fat which does not go into solution hence exerts no effect upon osmotic pressure and no restraining power over infection. From the standpoint of attack by molds, therefore, the significant percentage of moisture is total absolute moisture calculated against non fatty solids.

A few references to this literature involve Penicillia. *P. juglandis* Weidemann was described from moldy nuts in Germany. Fairman reports *P. crustaceum* as common in old, musty and decaying nuts in New York State.

*Almonds*

Kernel mold of almonds due to *Penicillium* was reported from the state of Washington by the Plant Disease Bulletin Suppl. 9, p. 161, May 15, 1926.

*Chestnuts*

Schell cites *Penicillium glaucum* with *Aspergillus niger* and *Carpocapsa splendens* as causing an annual loss of nearly one-third of the French chestnut crop.

*Filberts*

In his studies on moldiness in nuts Hartman states filberts tend to become moldy in the field unless they are gathered as soon as possible after falling to the ground. Immediately after gathering the nuts should be dried. Green filberts become moldy in a short time if kept in large piles or places where air circulation is poor. The water content of these nuts increases or decreases with the relative humidity of the air in which

they are kept. The water content of three varieties of filberts subjected to a range of humidities varied from 1.6 to 24.2 per cent. To be free from danger of mold filberts should be dried down to a water content of below 12 per cent.

#### OAT STRAW

Thaysen and Bakes noted *Penicillia* in chopped oat straw the water content of which was increased to 50 per cent by the addition of the requisite amount of water. The water used for wetting also contained sufficient ammonium carbonate to raise the nitrogen content of the damped straw to 2 per cent. The *Penicillia* appeared on the damp straw on the sixth day along with a very varied microflora, in and upon the straw packed in aerated boxes. Humification as related to this oat straw is discussed but not correlated to specific organisms.

#### OLEOMARGARINE

Mold colonies in the interior of "nut" butter were at least in two instances composed in part of growth of green *Penicillium*. *P. divaricatum* has been isolated by us from nut butter in four instances.

#### ONIONS

Hanzawa reported as parasitic a species with small greenish blue colonies, conidiophores about 290 by  $3.2\mu$  little branched, sterigmata in one-verticil up to 16 by  $3\mu$ , conidia 3.2 by  $2.4\mu$  fairly punctate. He listed it as *P. canum* Preuss. About the only grounds for the identification is common occurrence upon onions.

#### PALM LEAVES

Specially treated palm leaves prepared in California for decorative purposes became moldy from the pink or rosy organism (no. 4214) variously called *Penicillium* or *Gliocladium roseum*.

#### PASTE

One species of *Penicillium* grew in library paste to which was added 0.75 per cent of a 40 per cent solution of formaldehyde as a preservative but did not grow in paste containing 0.75 per cent phenol or even 0.35 per cent. The species was not determined. Our original culture (no. 34) of *P. divaricatum* (= *Paecilomyces varioti* Bainier) was found in an old pot of library paste covering the whole surface.

## RUBBER

Rubber produced upon the tropical plantations and shipped to the manufacturing countries often develops spots and areas of injury by mold colonies among them *Penicillia*. This has been studied by various authors (Paine, F. T. Brooks, Sharples, DeVries,) working especially in the Federated Malay States. Under the conditions of temperature and humidity encountered in rubber producing countries, control of mold is very difficult. Collection of latex involves gross contamination with microorganisms. Coagulation and drying under the best of conditions do not entirely eliminate mold growth. Subsequent handling involves many points at which moisture may enter the package and reestablish the activity.

Among the organisms active, *P. maculans* Sharples was isolated and described from such materials produced in the Federated Malay States and was reported to be associated with objectionable spotting of the product. Efforts to obtain authentic cultures of the species failed but by description it was one of the monoverticillate series. DeVries, after prolonged tests, decided that the presence of mold on the surface of sheet rubber was not a serious cause of deterioration within the ordinary period of storage. Nevertheless the losses in commercial value due to the moldy appearance are still great enough to stimulate experimental work upon prevention methods.

Various investigators (Stevens, Bishop and Greenstreet, Edwards, Brown) have studied methods of mold prevention in rubber. Sodium silicofluoride appears to have been the most satisfactory of the reagents used by these workers. Work upon control of molds is still in progress in the rubber producing regions.

Our own studies of rubber have been limited to one series of imported samples in which one monoverticillate *Penicillium* appeared among several molds chiefly *Aspergilli*. In a series of cultures recently contributed by Weir from the Federated Malay States. *Paecilomyces*, one divaricate *Penicillium*, and several *Aspergilli* were found. The existence of particular species of *Penicillium* destructive to rubber is not established by the evidence thus far put forward, but the presence of various *Penicillia* among the miscellaneous contaminants which injure the commercial appearance of the product is shown by the cultures examined and the reports already cited.

## SOIL

A list of species of *Penicillium* reported in cultures from the soil would practically cover the entire genus. Sopp described most of his

sixty species as found in soil. Dale sent us a series in the *Scopulariopsis* or *P. brevicaulis* group from English soil. In our own cultures and those submitted by various workers many of the common species occur repeatedly. As factors in the decomposition of organic matter any or all of those forms no doubt play their part although no one has established for them any special significance in the maintenance of soil fertility. Through all of the soil cultures studied, however, one series of related forms is constantly seen. A group of these was described by us (see Pratt) as the Soil *Penicillia* and are grouped in the taxonomic section of this book under the section, *Lanata-divaricata* Chapter XVII and in Chapter XVIII. Waksman in studying New Jersey soils made a special study of one of them and found it active in the decomposition of cellulose (no. 4789). These forms are constantly seen in cultures from earth and appear to be widely present in the soil itself.

Surveys of the molds appearing in culture plates from soil have been made from time to time for many years. In addition to the "soil series" already discussed, members of the series typified respectively, by *P. viridicatum*, *P. spinulosum*, *P. frequentans*, *P. lilacinum*, *P. luteum*, *P. rugulosum*, and *P. funiculosum* are frequent. Species of *Gliocladium* are common. The frequency of these species seems to indicate that they maintain themselves in the soil more or less independently of fresh supplies of decaying organic matter although the primary decomposition of such material must be regarded as their primary function rather than the final reduction of the material toward the status of food for green plants. Cultures of many of these forms occur in all studies of the tilled area but it seems doubtful if most of them are more than feebly active below the surface layers as indicated in the following experiments.

*Penicillia* were grown in test tubes of soil such as sandy loam, heavy clay and greenhouse loam. The original moisture content of these soil samples was varied through exposure, sterilization and added water. *Penicillium pinophilum* (no. 1) grew in heavy clay and in sandy loam to the depth of 5 cm. *Penicillium luteum* (no. 11) grew in sandy loam to the depth of 5 cm. with the production of perithecia to 2.8 cm. An undetermined monoverticillate species grew in sandy loam and heavy clay to the depth of 5 cm., but did not grow in greenhouse loam at 4.8 cm. (no. 4083 Cit., no. 4019.2). *Penicillium divaricatum* did not grow in greenhouse loam to the depth of 4.8 cm. *Penicillium africanum* grew to the depth of 5.5 cm., in greenhouse loam (no. 42). *P. notatum* (no. 2541) did not grow down into sandy loam and heavy clay, but fruited on the surface of the heavy clay. One *P. frequentans* strain no. 2467 grew in



sandy loam to a depth of 5 cm. Conspicuous conidial fruiting occurred practically to the limit of growth with the sandy loam, but only on the surface with the heavy clay.

Colonies with the morphology of *P. anisopliae* or *Metarrhizium* have been obtained occasionally from soil cultures in which they were not, readily, at least, traceable to the presence of diseased insects. These cultures produce olive green conidiophores partly in aggregates of varying size and shape, often covering considerable areas partly as short coremia or massed stalks or conidiophores perhaps 1 millimeter in height.

Abbott (1923, 1926) listed a series of species of *Penicillia* isolated from soils in Iowa and Louisiana and studied some of them in vitro mixed with soil and various fertilizers. One species, *P. luteum*, was reported as oxidizing sulphur; *P. funiculosum* acted definitely in rendering raw rock phosphate more readily soluble; all produced varying amounts of ammonia from dried blood and cottonseed meal.

Gilman and Abbott gave an extensive bibliography of soil fungi from which they collected references to all species described as soil organisms. They redescribed all species of *Penicillium* obtained and identified by them in culture and described as new species, *P. crateriforme* (near *P. rugulosum*), *P. restrictum*, *P. guttulosum*, and *P. vinaceum*. They transferred *P. divaricatum* (*Paecilomyces varioti* Bainier) to *Spicaria* and described an organism related to *P. lilacinum* as *Spicaria violacea*. Two species in the genus *Gliocladium*, *G. catenulatum* and *G. atrum*, were described as new.

#### SUGAR

Amons reported *P. glaucum* as common, *P. luteum* and *P. rubrum* as present but rare in Java upon spoiling sugar. Grove (O.) reported *P. glaucum* upon various sugar solutions and that in his experiment sucrose at 65 per cent checked its growth, although there was slight development in one month even at 70 per cent. Kopeloff and Byall followed the invertase activity of *P. expansum* from 10 to 70 per cent at which no further activity was reported. Maximum invertase activity was found between 50 and 60 per cent. Van der Bijl lists certain *Penicillia* as present in deteriorating sugar.

Townsend (1904) records finding mold hyphae in a sample of stored sugar from which he cultivated a strain of *Penicillium* (not identified) which sporulated in saturated solution of sucrose within six to eight days. Attempts to obtain this culture were not successful. No one has confirmed the results.

Our own studies of spoiling sugar and products carrying high concentration of sugar tend to eliminate the *Penicillia* from any significant rôle in such deterioration even though some damage in incidental or experimental cases is not questioned.

Kopeloff studying sugar deterioration inoculated his material with spores and found evidence of some invertase from the presence of conidia in solutions so concentrated that no growth of mold occurred.

#### TOBACCO

H. R. Jones studying moldy tobacco in England sent us strains of several series including *P. roqueforti*, a strain near *P. frequentans*, *P. chrysogenum*, *P. ochraceum*, and several of the biverticillate series. Kas in Czechoslovakia reported unnamed species of *Penicillium*. In our own samples slightly moldy or musty samples of various types of cigars and canned tobaccos, *Penicillia* have not been seen in microscopic examination. We doubt whether damage due to *Penicillia* is a significant factor in the deterioration of tobacco except when grossly mishandled.

#### WATER

Cultures from commercial bottled waters frequently show colonies of *Penicillium*. In this way *P. spinulosum* has frequently been found in both domestic and imported samples. Various asymmetriforms were collected but few of them identified. *P. commune* was found once in a Spanish water; *P. divaricatum* several times from Texas; *P. stoloniferum* from a Spanish water. Since the number of colonies seemed to be greater in bottled waters after long storage, a number of bottles which had stood undisturbed for a long time were opened without shaking. In such cases, pin point colonies were often seen floating upon the surface of the water, and producing abundant conidial masses. Study of the corks showed in many cases abundant growth of *Penicillia* and *Aspergilli*. These colonies probably furnished the principal source of the spores scattered through the contents of the bottles. Cultures were obtained both from fresh waters and samples which had been tightly closed for periods varying from a few months to four or five years.

One of the monoverticillate *Penicillia* was inoculated into both filtered and unfiltered water. Fruiting portions of a species obtained from a commercial water sample were inoculated into sterilized filtered water and also sterilized unfiltered water. These two samples of water were in glass-stoppered bottles. Microscopic examination of the surface of the central portion and of the bottom portion of the water showed no

germination tubes and no mycelium, although colonies were regularly obtained in culture. Since the corks in commercial samples occasionally show mold visible with the hand lens the experiment was repeated with cork-stoppered bottles. The corks used were new and sterilized with the sample of water. The bottles of water inoculated with mold were well shaken to insure thorough wetting of the corks and to simulate conditions of jarring in the transit of commercial waters. Microscopic examination of the water contained in the prepared corked bottles showed mycelium. The corks of the experimental bottles when examined with a lens presented mature, thriving colonies of the molds.

These experiments confirm the conclusions reached from examining commercial samples that when water containing viable spores is placed in corked bottles, the spores lodging on the bottom of the stopper find in the cork sufficient nourishment for growth, at least in the case of the species of *Trichoderma* and the monoverticillate *Penicillium* used. As a result, the bottle of water is still further contaminated with mold when the spores fall into the water from the base of the cork. Unsterilized bottles, unclean corks, or corks used a second time may bring about the same conditions as water originally containing mold spores. Bottles which have been used for soft drinks are at times known to be used next for bottling water. Such bottles as ordinarily handled contain remnants of the soft drinks and develop heavy contaminations with mold. This followed by poor washing and lack of sterilization prior to bottling results in mold contaminations of the contents.

#### WOOD

*See also fibers, paper pulp*

*Penicillia* have been studied as a cause of damage to the fiber and discoloration in wood by Hedgcock, by the forest pathologists and the forest products workers of the United States Department of Agriculture, by Larue, and others.

Brilliant shades of red, orange and yellow were found as a result of members of the biverticillate group.

Exhaustive studies by the forest pathologists do not find *Penicillia* greatly injuring the strength of the timber. The damage appears to be limited to discoloration. In the course of this work many species have been submitted for identification but none of them seem to be specific to wood studies or sufficiently limited to wood as to be listed as part of its specific flora.

## CHAPTER X

### PENICILLIA PATHOGENIC TOWARD MAN AND OTHER ANIMALS<sup>1</sup>

Penicillia have been reported occasionally in connection with diseases of man, certain domestic animals and insects. Part of these species have been found in the microscopic study of pathological tissues. Part of them have appeared in cultures made from such material, or from sputum, excreta, or directly from tegumental lesions. The occurrence of recognizable structures within specific tissues has established the presumption of agency in such cases as the strains of Scopulariopsis (*P. brevicaulis*) separated from infections of the nails (onychomycosis), and certain species found in lung sometimes referred to as "pseudo tuberculosis" (Greeley, various correspondents). The probability of pathological activity on the part of some species of the penicillate molds is thus presented.

Identification to group relationship let alone to species, of the Penicillia involved in most of the pathological reports and clinical diagnoses in the literature, is on the whole unsatisfactory if not impossible. Even a large acquaintance with common molds and mycological literature does not make such an attempt less difficult. As an illustration of the present status of this work, descriptions and figures of species of Scopulariopsis even though imperfect often suggest the genus, but hardly substantiate the specific and varietal designations assigned anew in each case. Some of the species in this group may be presumptively determined by the color of the colony, smoothness or roughness of conidia and length of sterigmata. Thus *S. brevicaulis*, *S. brevicaulis* var. *glabra*, and var. *alba* are more or less definitely identifiable. *P. brevicaulis* var. *intermedium* is not identifiable as a brown form with smooth spores, because examination of herbarium material by the authors proves the spores to be delicately rough when mature.

Certain molds apparently isolated from fairly characteristic lesions thrive also as saprophytes and when in laboratory substrata, present forms indistinguishable from strains of the same group isolated from strictly saprophytic situations. Establishment of causal relations between these strains and pathological conditions in man has been diffi-

<sup>1</sup> The material in this chapter was collected by Dr. Margaret B. Church.

cult or often impossible with the range of experimentation available. If these are parasitic, then some strains may have physiological qualities rendering them capable of causing disease and yet be morphologically indistinguishable from cosmopolitan saprophytes. Correspondingly whole groups of species constantly encountered in studies of the decomposition of waste material are put under suspicion by these findings.

The cultivation of molds by pathologists has commonly been carried out on media devised for pathogenic bacteria. Such media contain animal substances rather than the sugars, starches, and related substances which are especially favorable for most of these molds. A mold may grow on the substances of the animal body as a pathogenic organism or as a secondary factor in a pathological condition, but in such an environment it commonly either does not develop its fruiting structures at all or develops such structures differing in form and color from the usual saprophytic cultures upon which our records and descriptions have been drawn. In such lesions, conidial apparatus, if present at all, is very much reduced; vegetative hyphae in some species are very slender and delicate, in others become large, vesiculose or even tend to break up into single cells. However uniformly such structures may be produced, comparative culture under the conditions favorable to the usual morphology of *Penicillium* is necessary before we can place such species correctly within this group. Although certain species of *Penicillium* have been studied and described only as found in pathological materials, hence are unknown in culture, most of the forms isolated from such sources have assumed the structures characteristic of saprophytic species when transferred to culture media rich in fermentable carbohydrate.

Much diversity of view as to the part played by *Penicillium* may be seen throughout the literature. Jimenez in discussing the mycotic infections of Venezuela in 1926 pictures but does not discuss *Penicillium* as a causative agent. However uncertain their identifications, *Penicillia* are included among the pathogenic molds discussed by Castellani and Chalmers, *Scopulariopsis* specimens among the illustrative slides distributed by Pollacci-Nannizzi, and more or less extensive discussions occur in various chapters upon mycotic diseases. Few of these discussions are adequate, yet enough evidence has accumulated to warrant the scrutiny of pathological material for members of the group. For our purposes we have brought together an abstract of the literature in which *Penicillia* are reported.

Pollacci-Nannizzi have published in four fascicles, prepared slides of fungi pathogenic for man and animals. These preparations were first

distributed from Siena in 1922 and include slides of *Scopulariopsis*. Similarly, Von Höhnelt in his series of slides and specimens, included various species of *Penicillium* upon fragments of insects with manuscript names attached but no adequate descriptions nor data to establish any real meaning aside from their interest as curiosities.

Since there seems to be little logical connection between the various pieces of work cited, we have simplified the presentation by arranging the discussions of *Penicillium* found in human medical literature in alphabetical order of regions of the body involved. Reports of mycotic lesions in other animals will follow in alphabetical order to the animals involved. The species names used by the describers, with their descriptive material have been placed in the natural groups represented when possible, and the remainder put alphabetically among the unrecognizable forms in Chapter XXV.

#### PENICILLIA IN THE ALIMENTARY CANAL

Turesson isolated fungi from the feces of man and of higher animals in the zoological garden of Seattle. In studying thirteen men, he was only able to isolate one *Penicillium*, *P. divaricatum* (*Paecilomyces*). From a bear, he found *P. avellaneum*; from a buffalo, *P. luteum*; from a lizard, *P. notatum*; from a frog, *P. frequentans* (identifications were all by Thom). Turesson's results would seem to indicate that the conidia of the common species of *Penicillium* which are certainly consumed in very great numbers with the food, do not or only exceptionally survive the processes in the alimentary canal of the warm-blooded animals.

#### BLADDER

Salisbury is quoted as having isolated a *Penicillium* (*P. pruriosum*) from the bladder of a man (original article not seen by authors).

#### BLOOD

Salisbury, previous to 1883, reports *Penicillium quadrifidum* as recovered from the blood of patients suffering from erysipelas.

#### BRONCHIAL ASTHMA

Van Lecuwen (p. 62) states that 8

"The substances occurring . . . in grain infected with some fungi are certainly allergens, because animals and human beings who tolerate these substances on first contact gradually become sensitized. But these substances may certainly have a primary irritating action, since eight-day-old guinea pigs (from a

non-sensitized mother) may show definite symptoms on their first contact with them. And it is easy to understand that those persons whose mucous membranes are more permeable than normal suffer more from the primary action of these substances. In many cases these allergens must be compared, not with anaphylactogens but with toxins. They have a primary toxic action but they are also allergic."

In a later paper Van Leeuwen in reviewing his investigations on "climate allergens" states that *Penicillia* common in moist and damp houses form allergens to which many asthmatics are sensitive. Castellani reports a case of bronchomycosis due to *Penicillium* in a Serbian soldier in Macedonia during the World War. He states that the man had been wasting for two months, expectoration was muco-purulent, at times bloody, and a few mycelial threads were present. Potassium iodide in full doses acted well.

#### EAR

Siebenmann in 1889 reported *P. minimum* as developing as a membrane in the ear of a patient.

Chisolm-Sutton believed that otomycosis is more common than is usually suspected and that in consequence a correct diagnosis is not always made. He reported seven cases, five of which he stated were mycoses due to *Penicillium*. In the first case, culture from mycelial fragments and spores gave *Penicillium*; in cases 2, 4, 6 and 7, smears showed mycelia and fragments of *Penicillium*.

#### EYE

Liergard reports *Penicillium* as the cause of an ulcerated human eye; the organism was cited by Raymond and Parisot as *Scopulariopsis Koningi*.

#### FEET

Raymond and Parisot in 1916 recorded a commonly observed condition of soldiers feet termed "gelure des pieds." This condition is said to be an infection, aggravated by humidity, and caused by *Scopulariopsis koningii* Oudemans. The identification of this brownish gray strain was made by Vuillemin.

Mantelli and Negri report *P. mycetomagenum* as a parasite in a case of melanotic granuloma of the human foot at Turin, Italy.

#### GONORRHEA

*P. gonorrhoeicum* Hallier, in Flora, 51: 294, 300, fig. 9, 1868, is described as a penicillate form (Prap. nr. 342) assumed by *Cladosporium*

*gonorrhoeicum*. Hallier in the course of a five years' study of pathological material prepared over a thousand mounts. His drawings were evidently made from these mounts. This species name is to be discarded for lack of any accompanying description. Associated by Hallier with gonorrhea.

#### GRANULOMA

Pollacci described *P. Burci* as the agent producing experimental nodules. Our cultures received from Dr. Westerdijk and attributed to Pollacci (4876.23a) belong with *Paecilomyces* (see Chapter XXIII).

#### HAIR

According to Castellani and Chalmers, 1910, *Penicillium barbae* causes a chronic affection of the beard, mustache, and occasionally the axillae, presenting grayish or whitish punctiform formations. In natives of equatorial Africa and of Ceylon who do not bathe frequently, such as old persons and beggars, the skin presents dirt in which such fungi live saprophytically.

#### LUNGS

Castellani and Iacono report a case of a Serbian soldier diagnosed as tuberculosis, which was cured promptly with potassium iodide. A species of *Penicillium (crustaceum)* was isolated from the sputum of this case.

Giordano reports one case of a man in whom *Penicillium (crustaceum)* was the exciting factor. This case ended fatally when the man was thirty years of age. Filaments of mold were noted, using thionin as a stain and also pinhead masses of penicilli about the edges of lesions.

We are not entirely convinced that *Penicillium* even under suitable conditions is pathogenic. It seems probable that in those cases in which it has been found, it exists as a chance saprophyte in an already established lesion.

Greely examined sputa of fourteen cases of chronic non-tuberculous lung diseases and determined that these contained "*P. glaucum*." A vaccine of "*P. glaucum*" gave marked local and general reactions. Autogenous vaccine of the same organism given to a houseworker suffering with chronic bronchitis for twelve years cured this same patient. Greely and Brereton reported fourteen cases (perhaps the same as above) of chronic non-tuberculous lung disease from which "*P. glaucum*" was recovered. These strains of *Penicillium* grew at 70° to 80°F., but not at 98°F. These investigators claim that by using selective media various higher fungi as for example, "*P. glaucum*," may be cultivated from sputa



of such cases. These fungi were regarded as probably potent in the tubercular disease process, acting alone or with the tubercle bacillus.

Dr. Greely concluded that about 3 per cent of his patients clinically diagnosed as tuberculosis had been shown to suffer from *Penicillium* infections in the lungs. Observations of the same general nature have been verbally presented to us by members of the faculty of the University of Indiana School of Medicine. Forry (1921) in personal correspondence contributed a *Penicillium* near to *P. citrinum* which he believed significant in a case of this kind. Dr. Mary Lapham working the mountain region of our southern states reports many mold cases of this general description.

Stokes and McCleary in 1928 reported the autopsy of a fifteen year old colored boy who died from extensive tuberculous peritonitis and an acute fibrinous pleurisy of the lower right lung. In connective tissues of a bronchus adjacent to necrotic connective tissue *Penicillium* (or an *Aspergillus*) was found. Branching mycelia penetrated the necrotic bronchial wall and the peribronchial tissue. No cultures were made.

#### LYMPH VESSELS

Bruno Bloch reported a fungus later named *S. blochii* in gummatous lymphangitis having the clinical picture of sporotrichosis.

#### MYCETOMA

*P. mycetomi* Neveu-Lemaire: This name is given by Neveu-Lemaire (*Précis de parasitologie humaine*, p. 123, 4th edition, Paris, 1908) to a *Penicillium* which he claims both Brumpt and Bouffard noted independently. This *Penicillium* was recovered from a "mycetome du genou à grains rouges."

No real description was presented.

#### NAILS

Species of *Scopulariopsis* seem to be definitely related to mycotic infections of the nails (onychomycosis). Emile-Weil and Gaudin report *S. cinerea* in 1919 from one case where the fungus appeared as knotty filaments, terminal and intercalary "chlamydospores" and rarely as brown mycelia and conidia, and *S. brevicaulis* var. *hominis* from toes, where there developed chlamydospores and conidia. Sartory reports *S. brevicaulis* var. *unguis* in eight cases in 1919 and in one other case of onychomycosis *S. aureus* as irregular filaments, terminal and intercalary bodies and conidia. Brumpt and Langeron report two cases due to "*S. hominis*

*brevicaulis* var.," where mycelial filaments and chlamydospores were recovered from the toes.

#### SKIN

We have put here for lack of a better classification, the whole series of dermatomycoses and other lesions in which the source of a fungous culture is a break in the integument. Study of the reports together with many cultures we have received for identification as taken from such cases agree in pointing to contamination of a lesion due to some other agent as the source of the *Penicillium* spores producing the culture in many of these reports. Active agency of certain species of the true *Penicillia* is not excluded, while the accumulated evidence against the *Scopulariopsis* group indicates real responsibility. Among the probable contaminations are such species as *P. grande*, *Coremium syphiliticum* and *P. syphiliticum* of Hallier.

Castellani claims that species of *Penicillium* are correctly associated with pinte or carate. Scrapings from patches show, he states, generally a mycelium from which branches somewhat shorter and thicker originate at various points. These branches terminate in large penicillium-like fructifications. *Penicillia* may eventually be soundly connected with pinte or carate of Colombia and neighboring regions. Castellani claimed the form to which he gives the name *P. montoyai* is justly associated with pinte, but according to Menk of the Medical Department of the United Fruit Company in Santa Marta, Colombia, *Penicillium* does not seem to occur frequently or act as an important causative agent in carate. It was impossible to make out the presence of such fungi by microscopical examination of scrapings from the lesions of the skin, although mycelial elements and spores were noted in many cases. Menk in 1926 states that he has "never been able to find fructification forms that resemble *Penicillium*," in carate, while admitting that the varieties of fungi found in more than fifty patients examined at Santa Marta, Colombia, in three months and a half were not subjected to differentiation on culture media because rigid technique and any considerable experience with such methods was not possible. He thinks that mycosis is an associated etiological factor in carate.

Jannin, with the coöperation of Vuillemin, isolated *S. koningii* (Oudemans) Vuillemin from a subcutaneous gummata.

Castellani in 1925 differentiates black (including bronzine and blue) pinta into classes, one of which is characterized by patches of black or bronzine hyperpigmentation, occasionally with a bluish tinge. In

this class the only molds found were common saprophytes as *Penicillium*, etc., identical with those occurring on the skin of natives in the tropics under normal conditions. He regards the etiological rôle of such organisms as very doubtful.

Chavarria and Shipley in 1925 state that, while fungi from the red variety of pinta have in artificial media a somewhat typical penicillus, they believe this appearance is due to aborted growth of the fungi.

Neveu-Lemaire maintains *P. pictor* as a distinct species causing grayish violet carate but later reviewing in 1920 carate and pinta he does not mention *Penicillium*.

Frescoln of the United States (1916) Indian Service, pictures and mentions *Penicillia* as playing an important part in dermatology.

Leger and Nogue describe a green species as *Scopulariopsis leproides* which they report as the cause of dermatomycosis of the forearms and hands of two Musselmen who always struck the ground in salaaming hence came into frequent contact with soil. The description reads more like a *Cladosporium* than a *Scopulariopsis* (see Chapter XXVI).

Boucher reports strains of *Scopulariopsis* which he groups as *S. ivorensis* causing serious lesions localized over bony areas and leaving large, deforming cicatrices. He states that in Bassam and Bouake during 1915 to 1916, 33 per cent of his consulting cases were for mycoses. Such cases were rarely mortal, endured little suffering and did not generally require hospitalization. *Scopulariopsis* sp. were obtained from sixteen cases, fifteen by cultural technique. Microscopic examination of the blood from one case showed round cells,  $10\mu$  in diameter with thickened membrane,  $1\mu$ , and fluid from another tumor showed round, colorless, refringent, bodies,  $10\mu$  in diameter, with a thickened ornate wall. Microscopic observations are not given for other cases, and in the two cases reported the bodies are noted as rounded and not as truncate.

*Scopulariopsis ivorensis* as described by Boucher was obtained from subcutaneous lesions over bony areas as the cheek bone, arm, above the eye, in a young negress, leaving large, deforming cicatrices. Animal inoculations were undertaken using large and also young pigeons and guinea pigs. The fungus was generally pathogenic for the pigeons from which it was recovered in pure culture. In the guinea pigs there was no generalized disturbance nor anatomical lesions and the inoculated microorganisms disappeared.

Other strains were recovered from similar lesions over other bony areas. One other strain was inoculated into an adult and also a young pigeon. These birds were affected by the treatment but recovered in

four and a half to five months. Agglutination tests with two of the recovered strains were positive, one at 1/100 and the other at 1/50.

Boucher states that all strains studied by him were varieties of *S. brevicaulis* which he designates as *S. ivorensis*. This specific name may justly be retained if later medical investigations substantiate the relation of the organism to the particular type of lesions.

Sartory in 1916 in *Progres medicale*, notes a case of two gummatous lesions on the right thigh (cuisse) of a milkman due to *S. koningi*. These lesions were little, semifluctuating tumors about four and a half centimeters in diameter. Direct examination showed mycelia.

#### TONGUE

Panayotatou reported *P. "Linguae* (genre *Scopulariopsis*)"<sup>2</sup> as a species isolated from the infected tongue of two year old child, at Alexandria, Egypt. Glucose and mannite were favorable sources of carbon; optimum temperature 25° to 35°C.; serum was not liquefied; milk not coagulated; cultures developed well on neutral or slightly alkaline media, but poorly on acid media. Colonies became heavy wrinkled masses in shades of gray brown to brown after forty days. Cultures were not pathogenic to guinea pigs. A young cat inoculated at the base of the tail died in several hours from septicemia.

The descriptive notes fail to give any conception of the actual structure of this mold. The figures are absolutely uninterpretable. The vague suggestion of identification as a *Scopulariopsis* by Langeron is about the only tangible evidence given as to the genus involved. Unless actually purified and properly described by someone, the species must be discarded. The notes given follow: Cultures on Sabouraud's agar developing rapidly as a thin white mycelium forming a fine velvety surface becoming (verdatre) greenish and finally brownish, thick and wrinkled and adherent to the substratum, resembling the growth seen on the child's tongue; conidia in short chains apparently oval to spherical with a point at one end, 3 to 6 $\mu$  in diameter.

#### VAGINA

Salisbury is quoted as having isolated a *Penicillium* which he named *P. pruriosum* (original not seen by the authors) from the vaginal mucus in a severe case of pruritus.

Perrazzi reported *P. crustaceum*, *P. album*, *P. coccophilum*; and *P.*

<sup>2</sup> This is the actual form of the name given in the article.

*digitatum* from the vulva and vagina of a woman infected with puerperal fever. He found 33 per cent of the 260 swab examinations he made positive for common fungi.

#### VENEREAL GRANULOMA

Greco relates a *Scopulariopsis* which he names *S. venerei* to cases of venereal granuloma.

#### WHOOPIING COUGH

Hall discusses *Penicillium* in connection with what he terms "concealed" whooping cough. He used stomach lavage and thus brought up for examination white tenacious mucoid masses which consisted of "thin-walled semimucilaginous threads, 2 to  $4\mu$  in diameter, each divided into cells 10 to  $50\mu$  long, together with some round or oval spore-like bodies of the same diameter." The spore-like bodies were reported as developing into *Penicillia*. Six cases of whooping cough observed by Hall were in this fashion related to one species of *Penicillium*, one case was negative and a last case due to a different species of *Penicillium* was not cured by stomach lavage.

#### WRIST

Jannin reports a lesion on the dorsal side of the right wrist, consisting of a semi fluctuating tumor about 3 cm. high, 5 cm. long and 3 cm. wide in a girl of twenty years. A similar lesion was noticed at the same time on the teat of a cow in the stable of the farm where the girl lived. Jannin isolated *Scopulariopsis koningii* as the predominating organism from both lesions.

#### EXPERIMENTAL MYCOSIS

Meineri experimented with subcutaneous inoculation and inoculation on scarified areas, using *Penicillium glaucum* (?). His treatments were applied to eleven individuals afflicted with syphilis, eczema, tuberculosis, etc. He positively states that any dermatitis occurring was due to foreign substances inoculated with the *Penicillium* and to the dead fungus itself acting as a foreign body.

#### COW

Jannin isolated *Scopulariopsis koningii* from a cutaneous lesion on the teat of a cow.

## GUINEA PIGS

Segal reports isolating a species of *Paecilomyces* (our 4696) from guinea pigs inoculated with the virus of typhus fever. Out of twenty-six guinea pigs inoculated either subcutaneously or intraperitoneally with a saline suspension of this fungus, all but one reacted, eight dying. The symptoms of the inoculated guinea pigs were those of a pyrexia, resembling experimental typhus fever.

*P. Burci* of Pollacci as used in certain experimental inoculations and found pathogenic appears to belong here.

## HOGS

Creech of the United States Department of Agriculture, Bureau of Animal Industry, in 1926, isolated a strain of *Scopulariopsis brevicaulis* from a diseased hog.

## MICE

Otu and Komaya described (1924) *Scopulariopsis Castellanii* as pathogenic to white mice but not to guinea pigs. The original was received from Castellani in Ceylon.

## DESCRIBED FROM INSECTS

*Bee mycosis*

In the experiments of Fielitz, two strains of *Penicillium*, termed *P. glaucum*, gave negative results in brood chambers. Infection was obtained by hanging in the chambers honey combs on which the fungi had developed.

In 1925 Burnside isolated a species of *Penicillium* from a worker larva nummy dying of disease. Burnside also found *P. corylophilum* common on brood combs of bees. On dead bees, he found *P. cyclopium* in Maryland near the District of Columbia and in Minnesota; *P. glaucorugineum* Sopp in Somerset, Maryland; and a member of the *P. pallans* group on dead bees in Minnesota. His subsequent (unpublished) studies tend to regard the *Penicillia* as mostly saprophytic rather than as causes of disease. Identifications of these *Penicillia* were made by the authors.

*Cicadas*

Von Höhnelt reported *P. cicadinum* as pathogenic upon large singing cicadas. It formed areas white then blue green and finally olive. A

slide in the Von Höhnel collection at the Cryptogamic Laboratory at Harvard University represents Von Höhnel's original material.

### *Crickets and grasshoppers*

Delacroix described *Monilia penicilloides* from dead *Gryllus campestris*, in Clion (Indre), France.

Field crickets and grasshoppers (*Oedipoda caeruleum* and *Stenobothrus biguttulus*) were infected by spraying with a powdery culture of the mold distributed in water. When the insects were dead they were placed in a moist chamber on sterile filter paper and the mold with which they were infected developed on their bodies; after five to six days at 16° *Monilia penicilloides* did not develop on infected insects which died the first day, but only on those which survived for a longer period. Crickets were more susceptible than grasshoppers.

### *Phylloxera*

Baccarini (1908) reported a yellow *Penicillium* as abundant in the bodies of dead *Phylloxera*. Their actual significance in the destruction of these insects was not determined.

### *Drosophila*

Frobisher in studying the relations between a red torula and a strain of *Penicillium* "*glaucum*" pathogenic for *Drosophila melanogaster* observed that these two organisms produce a bright lemon-yellow color when grown together in cultures. Also he noted that the *Penicillium* changed the torula into a hard, tough colony, the cells of the torula seeming to be distorted. Frobisher states that *P. "glaucum"* kills *Drosophila melanogaster* by (1) matting the torula cells in the guts of the insects so as to form a more or less solid mass, which may block elimination or engorgement, and (2) by invasion of hyphae into the intestinal wall with binding of the mass to the wall.

## CHAPTER XI

### CLASSIFICATION AND SUBDIVISION

#### CLASSIFICATION

*Penicillium* is characterized and discussed here as a "form-genus" in the great aggregate of form genera commonly known as the Hyphomycetes in which the conidium-producing colony is the basis of a scheme of classification which is in large part a purely arbitrary device for the identification of organisms found. Although many authors have transferred *Penicillium* to the Ascomycetes, following Brefeld's classic description of an ascogenous form of *P. glaucum*, the practical needs of the worker with molds amply justify Lindau (in Rabenhorst I, Pilze VIII, p. 154 et seq., 1907) in retaining the genus among the Hyphomycetes. Among the ascogenous species already known, Sopp found a very different type of perithecium in his *Acaulium* from that described by Brefeld. The series of perithecial forms in the section *Biverticillium* Biourge (*Biverticillata-symmetrica* of this book) present still another type of ascogenous form. Until someone can establish genetic relations more closely our purposes are best subserved by keeping the group together.

Since the characterization proposed is designed to exclude from *Penicillium* in the stricter sense certain series of species already described as *Penicillia* but quite divergent in their characters, the abstract of groups presented is broadened to cover the genera *Scopulariopsis*, *Paecilomyces* and *Gliocladium*. This abstract follows Lindau in arrangement without reference to the natural or genetic factors involved. The figures taken from Lindau may assist those unfamiliar with the vast literature of the hyphomycetes in placing their forms properly.

#### Class FUNGI IMPERFECTI

*Diagnosis:* Fungi in which the filaments of the mycelium are septate, and which are classified on the basis of characters of asexual fructification and of vegetative mycelium.

#### *Key to the Orders*

- A. Conidiophores borne in a pycnidium..... Order 1.—*Sphaeropsidaceae*  
ex.—*Septoria* (fig. 15)



- B. Conidiophores borne on a thick stroma, or acervulus. . Order 2.—*Melanconiae*  
ex.—*Gloeosporium* (fig. 16)
- C. Conidiophores borne on a cobwebby or more or less compacted mycelium;  
but not on a stroma or in a pycnidium. . . . Order 3.—HYPHOMYCETAE

*Key to the Families of the Order HYPHOMYCETAE*

- A. Hyphae colorless or bright colored; not brown or black  
Family 1.—MUCEDINACEAE
- B. Hyphae dark colored or black. . . . . Family 2.—*Dematiaceae*
- C. Conidiophores aggregated into compact fascicles. . . . Family 3.—*Stilbaceae*
- Note:* Fascicles or coremia consisting of aggregated conidiophores, occur in certain species of *Penicillium*, in which the

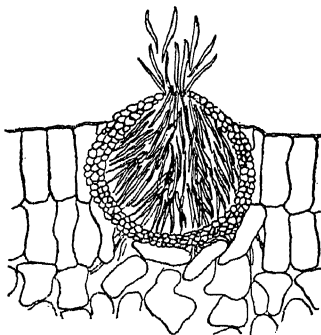


FIG. 15

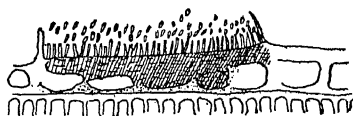


FIG. 16

FIG. 15. Diagrammatic section of a pycnidium of *Septoria* (after Lindau) as representing the structure found in *Sphaeropsideae*; showing its sac-like form with ostiole.

FIG. 16. Diagrammatic section of a sorus of one of the *Melanconiae* showing the dense mass underlying the fruiting area.

ordinary penicillate character is commonly evident in the same culture. The penicillate type of conidial fructification is present in certain species which always show the stilboid character. Certain of these species have been described as *Penicilli*, and are therefore discussed in this book.

- D. Hyphae compacted into a globose, discoid, or verruciform body or sporodochium; sporodochium typically sessile; waxy, or subgelatinous  
Family 4.—*Tuberculariaceae*

*Key to the Sections of the Family MUCEDINACEAE*

- A. Conidia single celled. . . . . Section 1.—HYALOSPORAE
- B. Conidia 2-celled. . . . . Section 2.—*Hyalodidimae*

- C. Conidia 3-or more celled.....Section 3.—*Hyalophragmiae*  
 D. Conidia 3 or more celled; spirally curved.....Section 4.—*Hyalohelicosporiae*  
 E. Conidia 3 or more celled; forked or star shaped..Section 5.—*Hyalostaurosporae*

*Key to the Sub-Sections of the Section HYALOSPOREAE*

- A. Conidiophores not sharply to be distinguished from the hyphae of the vegetative mycelium.....Sub-Section 1.—*Micronemeae*  
     ex. *Monilia*  
 B. Conidiophores readily to be distinguished from the hyphae of the vegetative mycelium.....Sub-Section 2.—*MACRONEMEAE*

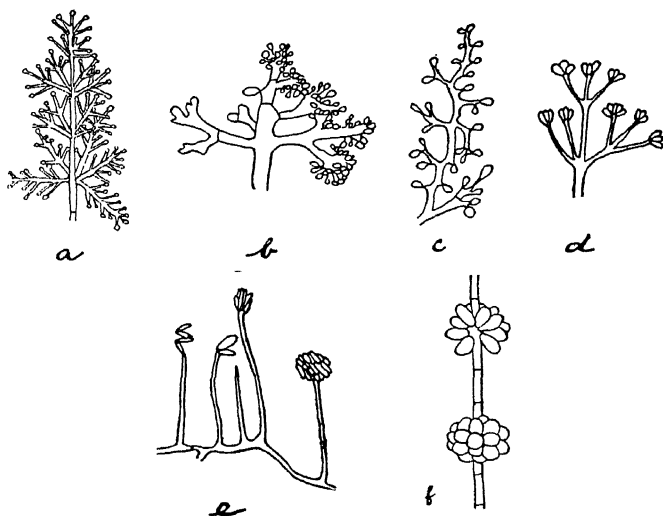


FIG. 17. Diagrammatic figures of Hyphomycetes (after Lindau): a, *Acrostalagmus*; b, *Botrytis*; c, *Sporotrichum*; d, *Verticillium*; e, *Cephalosporium*; f, *Gonatobotrys*.

*Key to the Groups of the Sub-Section MACRONEMEAE (fig. 17)*

- A. Conidia in heads.....Group 1.—*CEPHALOSPORIEAE*  
 B. Conidia borne irregularly on simple or branched, but not inflated or verticillate hyphae.....Group 2.—*Botrytideae*  
     ex. *Botrytis*  
     ex. *Sporotrichum*  
 C. Conidia acrogenous on verticillate branches.....Group 3.—*Verticillieae*  
     ex. *Verticillium*  
     ex. *Spicaria*  
     ex. *Acrostolagmus*

- D. Joints of the hyphae inflated here and there, and bearing pleurogenous conidia.....Group 4.—*Gonatobotryideae*  
ex. *Gonatobotrys*

*Key to the Subgroups of the Group CEPHALOSPORIAE*

- A. Conidia borne in heads, massed when ripe, not in chains  
Sub-group 1.—*Cephalosporiae*  
ex. *Cephalosporium*
- B. Conidia borne in chains.....Sub-group 2.—*ASPERGILLEAE*

*Key to the Genera of the Sub-Group ASPERGILLAE*

- A. Fertile hyphae inflated at apex, includes the genera  
*Dispira*, *Aspergillus*, *Dimargaris*
- B. Fertile hyphae little or not all inflated at apex
- c. Fertile hyphae verticillately branched in one or more series; conidia globose or elliptical, produced successively from special sterigmatic cells.
- Includes the genera *PENILLIUM*, *GLIOCLADIUM*, *SCOPULARIOPSIS*, and *PAECILOMYCES*

ACCEPTED GENERA

In seeking a scheme for systematic arrangement of the species already found in the *Penicillium* literature and for fitting the notes and observations from many thousands of cultures into this composite framework, the various generic usages proposed have already been considered. As a conclusion, such generic names as, *Scopulariopsis*, *Gliocladium* and *Paecilomyces* (already characterized, Chapter IV.) are probably useful enough and distinct enough to be tentatively retained. The needs of description and identification seem by the same tests to be better served by making the other subdivisions into subordinate groups under the generic name *Penicillium* as already characterized.

*Scopulariopsis and Gliocladium*

Removal of the *P. brevicaulis* series as *Scopulariopsis* is readily accomplished by examination of the conidium and its method of production combined with other and easily determinable characters since the group for the most part is one so homogeneous as to be readily recognized after one of them has been studied. Similarly the mucilaginous conidial masses of *Gliocladium* are easily recognized and may be used as a basis for segregation, although we know entirely too little about the species to define lines of real relationship among them.

*Paecilomyces*

Another section which may be separated contains a whole series of strains typified in our collection by *P. divaricatum* Thom, which is synonymous with *Paecilomyces varioti* previously described by Bainier. Corollium Sopp probably was based upon some member of this group, but we have not seen one of these molds with the large spores described for *C. dermatophagum*. The peculiar sterigma and the arrangement of sterigmata in this series are the only readily defined grounds for generic separation from Scopulariopsis and Penicillium. We failed to recognize Paecilomyces from Bainier's description and Biourge failed to recognize either Paecilomyces from Bainier's description or *P. divaricatum* from Thom's figures and description. It is, therefore, with much hesitation that we propose to use Bainier's generic name, even though observation of strains from many parts of America, from Europe, and from Asia indicate the cosmopolitan nature of these forms and their distinctness when once properly understood.

*Penicillium (more restricted sense)*

The great mass of Penicillia remain to be considered. Dierckx (1901) divided his series into two groups, Aspergilloides and Eupenicillium (not following Ludwig). He defined Aspergilloides to include Wehmer's genus Citromyces, rightly discerning that there is no sharp line between the monoverticillate forms and the other Penicillia. He did not go far enough, however, to indicate natural lines of relationship among the divergent members of either group. Bainier following Wehmer held to the separation of Citromyces from Penicillium, but did not find group relationships among the difficult lot of green Penicillia. Thom, in 1910, Westling in 1911, and Sopp in 1912, made no real progress toward indicating the lines of separation among these forms. Wehmer in 1914 and Thom in 1915 brought together the biverticillate series of species into the Verticillatae of Wehmer and the more restricted *P. luteum-purpureogenum* group of Thom.

## GENERIC SUB DIVISIONS

Biourge (1923) accepted Dierckx's two subgenera, offered the name Monoverticillium as a substitute for Aspergilloides and (Monog. p. 331) summarized in the form of a table of contents after his index, his scheme of classification as worked out in his presentation of species. This differs somewhat from the "résumé" given and discussed on his pages

28 to 33. The analysis on page 331 is presented here, using his Latin names for the subdivisions and leaving out his French synonyms:

I. Subgenus *Eupenicillium*.

Section *Bulliardium* or *Asymmetrica*.

Sub-section I. *Zonata* or *Concentrica*.

a. *Euzonata*.

b. *Hemizonata*.

Sub-section II. *Radiata* Biourge 1920.

Sub-section III. *Lanata*.

Sub-section IV. *Stellata*.

Sub-section V. *Fragilia*.

Sub-section VI. *Anomala*.

Section *Biverticillium* Dierckx 1900 (? date C. T.).

II. Subgenus *Monoverticillium* Biourge 1920—*Aspergilloides* Dierckx 1901.

A. series *Corylophilum* Dierckx.

B. series *candido-fulvum* Dierckx.

C. series *citrinum* Thom (*Thyrsifers*).

D. series *stipes acrocarp*, non-divisus.

Numerous difficulties arose in trying to use this classification. Biourge's subgenus *Eupenicillium* is made to include as "Sub-section VI. *Anomala*," the group of species typified by Saccardo's *P. brevicaulis* which was taken by Bainier followed by the French mycologists and others as the type species of the genus *Scopulariopsis* and is so accepted here. Biourge goes so far as to put *Stemonitis* in this section, thus introducing even greater divergence from the true *Penicillia* signified in the name *Eupenicillium*. Again he includes in *Eupenicillium* his section *Biverticillium* in which ascospore production, as studied by Zukal, Wehmer, Dangeard, Wortmann, Thom and Biourge himself, is in marked contrast with the method of ascospore production described by Brefeld for *P. glaucum* and Schwartz for *P. italicum*. Further comparative study will probably carry this section even to generic rank. Again our own studies indicate that his *Monoverticillate* type of penicillus shades into his *Eupenicillate* type so gradually as to weaken its value as a character in separating subgenera.

After having tried many provisional groupings we are proposing to drop the use of these subgeneric names with their taxonomic significance and group the species into four divisions using for names descriptive Latin adjectives adapted from the usages of Dierckx, Biourge and Zaleski. These and the sectional names proposed have been selected as far as possible for the purpose of making these names self-explanatory as chapter headings.

The four divisions proposed are: I. Monoverticillata, II. Asymmetrica, III. Biverticillata-symmetrica, and IV. Polyverticillata-symmetrica.

*I. Monoverticillata.* (Covering part or all of *Citromyces* Wehmer, *Aspergilloides* Dierckx 1901, and *Monoverticillium* Biourge.) The Monoverticillata constitute an aggregate of species and series of species bound together by the arbitrary fact that the penicillus in the division consists of a single cluster or verticil of sterigmata at the tip of a fertile hypha or conidiophore. This fertile hypha is usually unbranched but in those sections in which branching occurs the individuality of the terminal verticil of sterigmata each producing a chain of conidia, is maintained. Naturally this division of the genus includes the larger part of the species included by Biourge in his subgenus *Monoverticillium*, by Dierckx in *Aspergilloides*, by Wehmer, Bainier and Sartory, and others in *Citromyces*.

Very few species are absolutely monoverticillate—that is, produce conidiophores entirely without branching below the terminal cell which bears the verticil of sterigmata. Two sections of the division are however separable, the first designated “*Stricta*” (Chapter XII.) in which the fruiting hypha or conidiophore only occasionally shows a branch bearing a similar penicillus or shows a duplication of the Penicillus by a coordinate cell or metula from the uppermost septum. The second, *Ramigena*, (Chapter XIII) covers a series of species reported first by Bainier and Sartory and latterly by Zaleski in which the fertile hypha or conidiophore is nearly always branched at various septa with the branches of varying length, mostly diverging and not assuming the appearance of a characteristic brush or broom.

*II. Asymmetrica* (Biourge) adapted. In discarding Biourge’s use of the subgenus *Eupenicillium* the name *Asymmetrica* has been selected to cover the species found in his sub-sections, I. *Zonata*, II. *Radiata*, III. *Lanata*, IV. *Stellata*, V. *Fragilia*, and some of the forms assigned by Biourge and Zaleski to *Biverticillium* and even to *Monoverticillium*. In this usage the name *Bulliardium* although preferred by Biourge has been disregarded in favor of his own alternate term, *Asymmetrica*, which is descriptive of the general type of penicillus upon which the grouping is based. This penicillus contains terminal verticils of sterigmata bearing their conidial chains, supported by a secondary series of branches or specialized metulae usually arranged in a verticil asymmetrical or one-sided (lop-sided) or if symmetrical, subtended by one or more asymmetrically produced branches or groups of branches (incomplete

verticils) the whole forming the brush or broom commonly figured for *Penicillium*.

Every kind of variation is encountered among the penicilli observed in the various species assigned to the division but the asymmetrical penicillus with at least two series of elements, sterigmata and metulae, and commonly one or more subtending series of branches (rami, ramuli) is sufficiently characteristically present to be made the basis of this separation and the ground for our preference for the name *Asymmetrica* rather than *Bulliardium*.

*III. Biverticillata-symmetrica.* A coördinate division is created out of part of Biourge's *Biverticillium* (Wehmer's *Verticillatae*. The *luteum purpurogenum* group of Thom) for that series of species in which terminal closely packed verticils of lanceolate or acuminate sterigmata are borne upon symmetrically arranged verticils of metulae. More or less persistent ellipticity of conidia and yellow to orange or even bright red colors in sugar media are commonly associated with this morphology. Ascospores are known for such species as *P. luteum*, *P. petchii*, *P. avellaneum*, etc.

Wehmer (followed by Thom) discerned that while individual penicilli with this general structure are not uncommon in other groups, there is one whole series of obviously related species or strains in which this is the dominant type of penicillus. Occasional simplification, supplementary branching, or even triverticillate penicilli occur in these species but the combination of the biverticillate penicillus and the lanceolate sterigma of the group with the occurrence of the *P. luteum* type of ascosporic fructification only in certain strains of this same series, amply justifies the separation of this whole group from the other penicillate forms and its assignment to a coördinate division, designated as *Biverticillata-symmetrica* to separate it from a biverticillate but asymmetrical section, *Lanata-divaricata* in the division *Asymmetrica* already discussed and from the symmetrically polyverticillate division yet to be discussed. (For full discussion, see Chapter XX.)

*IV. Polyverticillata-symmetrica.* (*Synpenicillium Costantin* in part.) Costantin with a single species (*S. album* Costantin) under cultivation erected his genus (or subgenus, according to Bainier) *Synpenicillium* to cover penicillate molds in which the conidiophores branch off from superficial trailing, or ascending (not erect), and interlacing fascicles or ropes (funicles) of hyphae. This generic description would have brought together species of all three of the divisions already discussed into an unrelated group. Bainier justly saw these implications and

transferred Costantin's species back to *Penicillium* (*P. costantini* Bainier), but failed to recognize its real relationship to his own genus *Scopulariopsis*. Bainier then described several related species assigned by him to Costantin's *Synpenicillium* as a subgenus. These species when carefully scrutinized form a group sufficiently separate from the other *Penicillia* to raise a doubt as to whether they should be included at all. But since this review of the genus *Penicillium* undertakes to account for as nearly all the material assigned to the genus as possible, and since the information necessary for other disposal of these species is at present wanting, a coördinate division, *Polyverticillata-symmetrica*, is presented to cover Bainier's series of species in which the penicillus consists of a symmetrically polyverticillate (three to several series) branching system. Incidentally all described forms have their conidiophores borne from funiculose hyphae, and produce long elliptical conidia. These forms resemble each other in structure sufficiently to compel belief in a genetic relationship with perhaps *Scopulariopsis* as a more nearly related group than the true *Penicillia* (for full discussion, see Chapter XXI).

#### *Sub-divisions or Sections*

Within these divisions the numbers of species described are still so great that subdivision is necessary. In seeking lines for such subdivision part of the names proposed by Biourge have been used with some adaptation of limits, certain natural sections of the group are defined and arbitrary sections created for the rest. At best many described species and certain organisms in culture are insufficiently known to claim completeness or permanency for such subdivisions. They are only offered as a means of placing the species already carefully studied in as nearly natural relations as our present information will permit with arbitrary provision for those less adequately known.

The various sections are designated either by the names of important key species or by Latin adjectives descriptive as nearly as possible, of some major character used in separation. To avoid unnecessary repetition the full discussion of these sub-divisions is largely deferred to the sectional discussion. The abstract of the whole scheme of grouping employed, follows:

#### *Summary of divisions, sections and sub-sections*

*Genus Penicillium* (characterized in Chapter I).

- I. *Monoverticillata* (*Monoverticillium* Biourge, 1923, in part and *Aspergilloides* Dierckx, 1901, at least in part). Penicilli typically monoverticil-



- late either borne upon branched conidiophores or upon branches which are so arranged as to preserve the identity of each monoverticillate penicillus.....Chapter XII, XIII
- Section 1. *Monoverticillata-stricta*: branching of conidiophore when present, occasional, irregular and lacking the suggestion of a characteristic terminal verticil.....Chapter XII
- Sub-section 1. *Sclerotigena*: sclerotium producing species.
- Sub-section 2. *Floccosa*: colonies floccose.
- Sub-section 3. *Funiculosa*: funicles or ropes of hyphae consistently found among the aerial hyphae in the colony. Species of the sub-section begin to appear among floccose types and bridge the gap between sections 2 and 4.
- Sub-section 4. *Velutina*: colonies velvety in appearance.
- Section 2. *Monoverticillata-Ramigena* (see Chapter XIII): fertile hyphae (conidiophores?) prostrate, ascending or erect, mostly branching, with branches 1-celled (metula-like) or long and septate, simple or occasionally again branched, each bearing a terminal monoverticillate penicillus, but not producing apical verticils of metulae or branchlets.....Chapter XIII

## II. *Asymmetrica*.

Penicillus consisting of two or more series of elements including sterigmata and metulae with or without branches (rami) of one or more series, with the branching system typically lopsided, one-sided, or asymmetrical.

The groups of elements in such penicilli develop ordinarily in basipetal succession—the first group of sterigmata upon the tip of the main axis, a verticil of metulae from the next lower node or septum, and lower branches when present successively lower down on the conidiophore and incompletely verticillate or more or less one-sided in arrangement.

This general type of penicillus as described is an arbitrary common character bringing together several more or less natural groups of species and many single species whose affinities are very little known. General colony characters and more or less minor morphological differences are made the basis for sections and subsections.

- Section 1. *Velutina*: See Chapter XIV. Colonies velvety in appearance; strictly covering species with aerial portion consisting of conidiophores standing like a field of wheat but since many gradations are found, it is interpreted to include species with a basal network of aerial hyphae when those species are clearly related to species of the more strictly velvety type.

Three subsections (nos. 1, 2 and 3) contain species with conidiophore walls smooth or nearly so, that is, not conspicuously pitted or roughened. The other three sections (nos. 4, and 6) show conidiophore walls mostly conspicuously pitted or roughened.

- A. Sub-sections with conidiophore walls smooth.

Sub-section 1. *Velutina-elliptica-magna* (*Fragilia* of Biourge): Colonies known in culture, or described as, velvety, with conidia elliptical usually more than  $4.5\mu$  in long axis, desig-

- nated by Biourge as *Fragilia* on account of the ephemeral structure of the adult colony in *P. digitatum*.
- Sub-section 2. *Velutina-divaricata*: Colonies velvety; penicilli with metulae and ultimate branchlets divaricate so that each verticil of sterigmata stands out like a monoverticillate penicillus.
- Sub-section 3. *Radiata* (*P. chrysogenum* series): Colonies velvety, regular in outline, azonate or less commonly zonate at the margin in age; conidiophore walls smooth.
- B. Sub-sections with conidiophore walls pitted or rough.
- Sub-section 4. *Velutina-restricta*: Colonies forming narrowly growing compact masses.
- Sub-section 5. *Stellata* of Biourge (the *P. roqueforti* series): Colonies broadly spreading, mostly plane, with irregularly stellate margin, wide, during the growing period cobwebby (arachnoid) in appearance from coarsely radiating hyphae with alternate submerged and aerial segments, and with irregular development of interlacing branches.
- Sub-section 6. *Velutina asperula*: Colonies plane, broadly spreading, with margin regular or nearly so without the arachnoid appearance of the *Stellata* and differing from the *Radiata* in having rough walled conidiophores.
- Section 2. *Brevi-compacta* (see Chapter XV): Colonies varying from floccose with a tendency to a velvety margin in age, to almost velvety appearance in different species and under changing conditions at times in the same species; penicillus consisting of sterigmata, metulae, and commonly one or more branches closely compacted at base, diverging at the apex with chains of conidia parallel, diverging or tardily somewhat tangled, but not in columns. This section bridges the gap between *Velutina* and *Lanata* but is arbitrarily brought together by the characteristic penicillus described.
- Section 3. *Lanata-typica* (see Chapter XVI): Colonies floccose or lanose (hyphae not combined into trailing ropes or erect fascicles) developing as floccose felts within which conidial areas are produced; penicilli forming the compacted or complex brush or broom of the asymmetrica.
- Section 4. *Lanata-divaricata* (see Chapter XVII): Colonies lanose usually producing trailing or ascending (not commonly erect) aerial hyphae with terminal penicilli (asymmetrically biverticillate) consisting of a one-sided verticil of diverging mostly unequal metulae (or branchlets) bearing verticils of sterigmata and giving the effect of a cluster of monoverticillate penicilli.
- Section 5. *Funiculosa* (see Chapter XVIII): Colonies floccose in general appearance but under the microscope seen to have part of their aerial hyphae combined into trailing, branching, and usually anastomosing ropes or funicles (not into erect coremia), with conidiophores partly rising directly from submerged

mycelium, partly terminal upon or borne as branches from the ropes of hyphae; penicilli either divaricate as in *Lanata-divaricata* or compact as in *Lanata typica*.

Section 6. *Fasciculata* (see Chapter XIX): Colonies with part to all of the conidiophores aggregated into more or less definite and erect bundles, fascicles or coremia; penicilli asymmetrical; walls of conidiophores mostly pitted or rough.

A. Sub-sections showing fascicles and simple conidiophores usually closely aggregated in culture; simple conidiophores usually predominating.

Sub-section 1. *Sclerotigena*: arbitrary segregation of sclerotium producing species.

Sub-section 2. *Aeruginosa*: conidial areas in strongly blue green colors.

Sub-section 3. *Viridicata*: conidial areas in bright green to yellowish green shades.

Sub-section 4. *Glaucia*: conidial areas in dull green or only transiently bluish green, then green or dull yellowish green.

B. *Coremia* predominating and characterizing the colony.

Sub-section 5. *Coremiella*: colonies in which fasciculation is fairly strongly marked throughout the colonies.

Sub-section 6. *Coremium*: colonies characterized by erect and usually separate coremia.

III. *Biverticillata-symmetrica* (see Chapter XX): penicillus typically consisting of one symmetrical verticil of metulae, bearing symmetrical verticils of sterigmata, lanceolate or acuminate at apex.

Section 1. *Ascogena*. The species producing perithecia and ascospores are put together as the *Ascogena*. One sclerotium producer is arbitrarily included. In making this aggregate *P. avellaneum* Thom and Turesson, and *P. spiculisporum* Lehman whose affinities are doubtful have been included with a truly homogeneous series including *P. luteum* and its allies so recently discussed by Derx and Biourge. The non-ascosporic members of the *P. luteum* series are arbitrarily put together in Section 3 for discussion even though continuation of Derx's studies will probably ultimately show forms described separately there to be haplonts of some ascosporic species to be determined later.

Section 2. *Coremigena*. Species like *P. duclauxi* Delacroix with abundant and erect coremia are sufficiently differentiated to form an arbitrary but useful section in such a classification as this.

Section 3. *Luteo-virida*. Colonies commonly showing green conidial areas on mycelium more or less yellow to red.

Sub-section 3a. *Funiculosum* series. The species producing trailing and anastomosing ropes of hyphae have so much of morphology and biochemical reactions in common as to warrant segregation under the name of one of them as a series. Reverse and substratum are commonly yellow orange or red, in shades or successions characteristic of the strain or species.

Sub-section 3b. *P. luteum-purpureogenum* series. Colonies with conidial areas some shade of green, commonly bordered or inter-mixed with areas yellow from hyphae covered

with yellow granules which may become red in age; reverse and substratum yellow, to orange and often to red, differing in the intensity, rate of development and final shade of color attained.

Section 4. *Miscellanea*. The biverticillate type of penicillus is characteristic of a number of species part of which lack the typical sterigma and others lack the color reactions of *P. luteum* and its allies. These species have been aggregated into a subsection of this group for taxonomic and descriptive purposes. The members of the section are not regarded as genetically related and some of them show little in common with the other biverticillate sections.

IV. *Polyverticillata-symmetrica*: *Synpenicillium* Vuillemin in part (see Chapter XXI). Penicillus symmetrically polyverticillate: the branching systems consisting of three to several superposed series of (étages) verticils forming a fairly symmetrical mass; conidiophores borne as vertical branches from a network of prostrate or trailing branches and ropes of hyphae.

Genus. *Gliocladium*. Corda (for discussion see Chapter XXII).

*Gliocladium* Corda was described as reproducing the growth habits, mycelium, conidiophores and conidial apparatus of *Penicillium* except that the conidia borne successively from the tips of sterigmata become enveloped in mucilaginous drops which increase in size with the increased numbers of conidia, followed by the fusion of the masses upon adjacent sterigmata, then often the fusion of these mucilaginous masses with those from adjacent penicilli to produce large balls of conidia.

Matruchot has described perithecia and ascospore formation in certain species, but the forms constantly encountered in culture are purely conidial. Comparative studies of structure in both conidial and ascosporic forms are necessary before *Gliocladium* and *Penicillium* or its ascosporic sections can be safely placed with reference to each other among the Ascomycetes.

Genus. *Scopulariopsis* Bainier (for discussion see Chapter XXIII).

Genus. *Paecilomyces* Bainier (for discussion see Chapter XXIV).

Genus described as related to *Penicillium* and *Aspergillus*, distinguished from these genera by its sterigmata which are short tubular or more or less enlarged, tapering into long conidium-producing tubes mostly curved or bent slightly from the axis of the sterigmata; sterigmata occur variously, partly in *Penicillium*-like verticils with conidium-bearing tubes and conidial chains divergent, partly variously arranged on short branchlets, or again occurring singly and laterally upon fertile hyphae, in terminal groups often approximating the appearance of a *Penicillium*; conidia elliptical.

Bainier failed to describe accessory structures which appear more or less commonly in all species, very abundantly in certain species of this group. These are regarded as macrospores by Horne and Williamson and made the basis of transferring these species to *Eidamia* (see discussion Chapter XXIII).

## CHAPTER XII

### MONOVERTICILLATA-STRICTA

#### MONOVERTICILLATA

The monoverticillate *Penicillia* include the subgenus *Monoverticillium* Biourge (Monograph, p. 331), and its synonyms, *Citromyces* Wehmer, 1893, and *Aspergilloides* Dierckx, 1901.

#### *Group diagnosis*

*Penicillus* or conidial apparatus typically monoverticillate (fig. 18): the main axis of the conidiophore terminates in a more or less enlarged apex, like the vesicle of *Aspergillus*, which bears a cluster or verticil of sterigmata each producing an unbranched chain of conidia: (a) in occasional species commonly, and in many forms occasionally, a secondary fertile branch produced at the uppermost septum or node diverges to bear a second conidial mass; (b) in others, a trailing or ascending fertile hypha bears a terminal conidial apparatus and produces branches usually one or a verticil of divergent branches at the first node or septum with or without other branches at several lower nodes or septa, each bearing a terminal penicillus. Branches when produced are commonly unequal in length and irregular in occurrence, number and arrangement, and so divergent that the terminal penicillus upon each retains its individuality.

The name *Citromyces* proposed by Wehmer was based upon two particular citric acid producing cultures before he knew the wide range of species which would be included in the morphological characters proposed for this genus. Wehmer's description of his genus assumed that the conidiophore was always unbranched, hence produced a single apical verticil of sterigmata. On this basis the line between *Citromyces* and *Penicillium* was sharp. Actual study of organisms in culture, however, quickly showed that branching conidiophores appeared at least occasionally in colonies of nearly every species. There was seen to be a complete gradation between forms strictly monoverticillate, forms in which an occasional irregularly produced branch bore a second penicillus, and, forms in which characteristically clustered branches each bore the same kind of a penicillus, toward forms in which a complex

branching system largely obliterated the individuality of the single penicillus. Sopp and Bainier, however, continued to use the name *Citromyces*. Dierckx created his subgenus *Aspergilloides* for the whole group of forms. Westling and Thom merely dropped the name *Citromyces* and called all of them, species of *Penicillium*. Biourge used the name *Aspergilloides* in his discussion on page 31 and proposed the subgenus *Monoverticillium*. He made the characterization broad enough to include all species in which the individuality of the ultimate verticil of sterigmata was maintained even though its stalk had become a part of a fairly definite branching system.

Although the marks of division are more or less unsatisfactory, the individuality of each verticil of sterigmata with their conidial chains

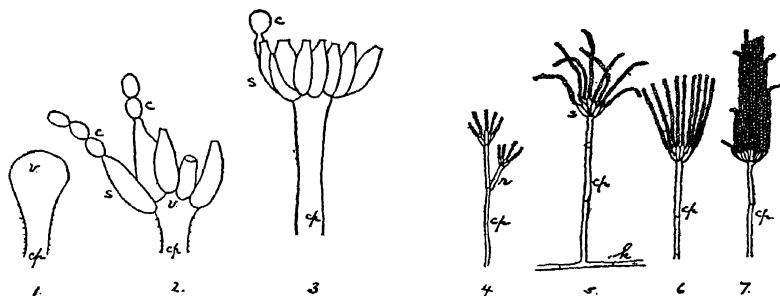


FIG. 18. The monoverticillate penicillus: *c*, conidia; *cp*, conidiophore; *h*, hypha bearing the conidiophore; *s*, sterigmata; *v*, vesicular apex; 1, upper end conidiophore with vesicular apex; 2, a verticil of sterigmata with tips diverging; 3, a verticil of crowded sterigmata; 4-7 diagrammatic representations of types of monoverticillate penicilli; 4, the ramigenous form with one branch showing; 5, the divaricate form with tips of sterigmata and chains diverging; 6, type with chains parallel; 7, type with chains adherent into a column.

is a character usually readily recognized when a growing culture in a petri dish is viewed with low magnifications of the compound microscope. For purposes of definiteness in our discussion separation between the monoverticillate group and the remainder of the *Penicillia* is based upon examination of the terminal verticil of the main conidiophore at the edge of a growing colony. If there are no branches at the apex of the main axis, or if each branch maintains fairly definite individuality, as producing a separate penicillus, the species is assigned to the monoverticillata. Necessarily arbitrary allocations occur in border series or subsections.

No analysis of the monoverticillata has yet assigned them all to groups or series showing real relationship. The sections and subsections proposed are, therefore, often arbitrary separations in which series with well defined relationship are aggregated with unrelated forms which merely have a few characters in common.

The general unsatisfactoriness of nomenclature among these species is well illustrated in the discussion by Pollacci who reduced all the species studied by him to synonymy with one aggregate species as *P. pfefferianum* (Wehmer) Pollacci.

*Key to species included in the Monoverticillata*

Sub-section 1. Sclerotigena

- I. Sclerotia not reported.....III
- I. Sclerotia reported.....II
- II. Conidia elliptical.....a and b.
- II. Conidia globose.....c, d, e, f.
- IIa. Conidia 3.5 by 2 $\mu$ . Sclerotia abundant pink...*P. thomii* Maire, 1.
- IIb. Conidia 3 to 3.5 by 2 to 2.8 $\mu$ .....*P. turbatum* Westling 67.
- IIc. Conidia globose, 4 to 5 $\mu$  in long axis. Clumps of sclerotia on potato.....*P. deformans* Sopp. 570.
- IIId. Conidia 5 by 7 $\mu$  (probably error), giving red color to colony; mycelium a tough white felt, "red" from sclerotia in reverse.....*C. alborosum* Sopp, 4.
- IIe. Conidia about 5 $\mu$ , steel-blue to brown, sclerotia only on colostrum. Soil.....*C. coeruleus* Sopp, 5.
- IIIf. Conidia about 2.5 to 3 $\mu$ , sclerotia doubtful *C. griseus* Sopp, 48.

Sub-section 2. Stricta-floccosa

- III. Colonies floccose—with more or less extensive development of aerial mycelium as simple (not rosy or fascicled) hyphae.....Stricta floccosa IV.
- III. Colonies with part of the aerial hyphae as ropes or funiculose masses or networks of trailing or ascending hyphae.....Stricta funiculosa XX.
- III. Colonies velvety in appearance.....Stricta velutina XXX.
- IV. Colonies in neutral gray to almost black tones, never green.....V
- IV. Conidial areas some shades of green or blue green during the growing period.....VI
- V. Reverse uncolored; conidia rough; 2 to 2.5 even to 3 $\mu$ .....*P. restrictum* Abbott, 6.
- V. Reverse orange; conidia rough 3 to 3.5 $\mu$ .....*P. echinatum* Dale,

- VI. Conidia larger than  $4.5\mu$  in long axis.....VII.  
 VI. Conidia less than  $4.5\mu$  in long axis.....X.
- VII. Reverse or substratum red.....VIII.  
 VII. Conidia large; reverse or substratum not  
 reddened.....IX.
- VIIIa. Conidia globose, echinulate, about  $6\mu$ d.,  
 occasional secondary heads. Felted my-  
 celium and red drops.....*C. purpurrescens*  
 Sopp, 10.
- VIIIb. Conidia brownish 5 by  $4\mu$ . Felted, buckled,  
 colonies blue green to brown.....*C. rubescens* Sopp,  
 11.
- VIIIc. Colonies olive to bluish green, with a tough  
 leathery mycelium, rosy to red or orange  
 red below; conidia 3 to  $4\mu$  in diagnosis but  
 described as coarse and echinulate and  
 given as 4 to  $7\mu$  in the table.....*C. virido-albus* Sopp,  
 12.
- IXa. Conidia smooth 6 by  $5\mu$  almost black abundant. *C. olivaceus* Sopp,  
 15.
- IXb. Conidia echinulate 5 to  $6\mu$ , brown.....*C. fuscus* Sopp, 16.
- IXc. Conidia 5 to 6 by  $4\mu$  in solid columns; conidio-  
 phores 30 to  $100\mu$  long.....*P. gliocladiodes*  
 Speg., 586, XXV.  
 See also Velvety.
- X. Conidia elliptical<sup>1</sup> (less than  $4.5\mu$  in long axis)...XI.  
 X. Conidia globose (less than  $4.5\mu$  in long axis)...XII.
- XIa. Mycelial mats dense rosy; reverse bright red;  
 conidia scanty 3.5 to 5 by 2.4 to  $3.5\mu$ .....*P. roseo-cinnabarium*  
 Biourge, 58, XII.
- XIb. Mycelium reddish to chamois red; conidia 3  
 to  $4\mu$  (3 to  $7\mu$  in Sopp's key). See VIIIc.....*C. virido-albus*  
 Sopp, 12, XII.
- XIc. Colonies green or bluish green; red color  
 abundant on potato, carrot, etc.; penicilli  
 terminal or on branches, vesicles up to  $8\mu$ ;  
 sterigmata few divergent; conidia 3 by 2 or  
 $2\mu$ .....*C. musae* Bainier and  
 Sartory, 96, XIII.

<sup>1</sup> These species by description have elliptical conidia, floccose mats of mycelium and reverse red but are placed elsewhere on details of morphology.



- XId. Colonies bright blue, floccose to hirsute with conidiophores branching from long ascending hyphae; conidia 4 by  $2\mu$  in columns. . . . . *C. cyaneus* B. and S., 95, XIII.
- XII. Conidia globose, less than  $4.5\mu$  in long axis. . . . .XIII.
- XIII. Colonies reverse colorless or partially rose in some species; without colored drops. . . . .XV.
- XIII. Colonies with reverse, agar, drops or all three in red shades. . . . . *P. sanguifluus* Series XIV.
- XIV. *P. sanguifluus* series.
- XIVa. Mycelium tough, leathery, wrinkled, with red drops abundant; conidia  $1.5\mu$  ( $3\mu$  in key) chains parallel or divergent; conidiophores very short branches of aerial hyphae. . . . . *C. sanguifluus* Sopp, 17.
- XIVb. Mycelium or felt wrinkled or buckled in quadrants, 200 to 400 thick, hyphae  $2\mu$ , conidiophore less than  $100\mu$ ; conidia about  $2\mu$ . . . . . *P. roseo-purpureum* Dierckx, 18.
- XIVc. Probable synonym: blue green to gray blue; rose areas and rose colored drops; conidia  $2\mu$ . . . . . *C. cesiae* B. and S., 19. *C. sanguifluus* Sopp.
- XIVd. Colonies blue green to fuscous or ashy gray, mycelium a floccose felt, close textured, wrinkled and faintly zonate; reverse yellow, orange, then brick red; conidia about 2.8 to 3.2, given also as 2.5 to 3.5 by 1.8 to 2.4. Compare. . . . . *P. subluteritium* Biourge, 89, XII.
- XIVe. Red color limited to a pink or peach coloration or margin. Colony thin, little floccosity, bluish green then pale green or gray green (olive); conidial branches tend to be clustered at the ends of creeping hyphae as well as lateral; conidia  $2.8\mu$ . . . . . *Cit. ramosus* B. and S., 101, XIII.
- XIVf. Colonies forming broadly spreading, cottony masses up to 5 mm. deep, slowly becoming gray green CDC 346; reverse rose but no color excreted; penicilli borne on short lateral branches; sterigmata few 5 to 6, and up to 8.4 in length, divergent; conidia up to  $2\mu$  globose (soil series?). . . . . *C. minutus* B. and S., 97, XIII.

- XV. Reverse colorless, or pale shades of yellow-lowish even rose.....XVI.
- XVI. Conidia globose or subglobose, more or less spinulose.....XVII.
- XVI. Conidia globose or subglobose, apparently smooth otherwise as in XVII.....XVIII.
- XVI. Colonies probably belonging here by general accepted discussions.....XIX.
- XVII. *P. Pfefferianum series.*
- XVIIa. Colonies with a loose, mostly prostrate network or aerial hyphae bearing most of the conidiophores which form a loose rather than compact conidial area, dull green, with white areas of mycelium exposed; conidia subglobose spinulose 3.2 to 3.5 by 3.6 to 4 $\mu$  or subglobose 3.5 to 4 $\mu$ .....*P. spinulosum*, 25.
- XVIIb. Colonies dark bluish green; reverse yellowish or reddish; conidia 3 to 4.5 granular....*P. flavo-cinereum* Biourge, 26.
- XVIIc. Colonies bluish green to chocolate brown in age; conidia 2.6 to 3.5 $\mu$  granular.....*P. fluitans* Tiegs, 27.
- XVIIId. Colonies dark gray green; conidia 3 to 4 $\mu$  spinulose.....*P. Pfefferianus* in Biourge, 28.
- XVIIe. Colonies deeply lanose, dull green to dark gray green; reverse reported by Biourge as violaceous; conidia faintly spinulose 2.5 to 3.5 $\mu$ .....*P. roseo-maculatum* Biourge, 29.
- XVIII. Conidia reported as smooth or without record of surface.
- XVIIIa. Conidia greenish to gray, 2.3 to 2.8 $\mu$  smooth....*C. Pfefferianus* Wehmer, 31.
- XVIIIb. Reverse rose, conidia 3 to 3.5 smooth.....*C. brunziai* Sartory, 33.
- XVIIIc. Colonies glaucescent; conidia smooth 2 $\mu$ .....*C. sormanii* Carbone 34.
- XVIIIId. Colonies blue green to green, to dark gray shades; reverse in pale yellow or orange shades, to dark brown conidia 2.5 to 3.5 $\mu$  smooth.....*P. Trzebinskii* Zal, 35.
- XVIIIe. Colonies about 500 $\mu$  deep, almost velvety especially at margin and extending in thin radiating lines; conidial areas in gray to

dark green; conidia 2.5 to 3 or even 3.5 $\mu$   
 smooth or with double wall and doubtful  
 development of wrinkles or spinulosities....*P. viridi dorsum* Bi-  
 ourge, 30.

XIX. Probable members of the series but inade-  
 quately described:

- XIX. ....(a) *C. citricus* Mazé  
 and Perrier, 36.  
 XIX. ....(b) *C. lacticus* Mazé  
 and Perrier, 37.  
 XIX. ....(c) *C. oxalicus* Mazé  
 and Perrier, 38.  
 XIX. ....(d) *C. tartricus* Mazé  
 and Perrier, 39.

### Sub-section 3. Stricta-Funiculosa

- XX. Colonies showing part of their aerial hyphae  
 as branching and anastomosing ropes.....XXI.  
 XXa. Colonies grayish or with a trace of violet  
 tinge scarcely any green.....See XXIII *Carmino-*  
*violaceum*, 40.  
 XXI. Ropiness or funiculose condition well marked  
 as the colonies are examined under the  
 microscope.....XXII.  
 XXI. Ropiness reduced to trailing and more or less  
 fascicled hyphae in colonies velvety or  
 nearly so in appearance.....XXVI.  
 XXII. Conidia elliptical.....XXIII.  
 XXII. Conidia globose.....XXIV.  
 XXIIIa. Colonies scarcely or very transiently green-  
 ish, restrictedly growing, forming wrinkled  
 or buckled felts, yellow to orange to violet  
 red yellow.....*P. carmino-violaceum*  
 Biourge, 40.  
 XXIIIb. Colonies closely felted, greenish white; re-  
 verse sordid orange, rose to red or almost  
 black.....*P. chermesinum* Bi-  
 ourge, 41.  
 XXIIIc. Colonies green to gray green and finally  
 brown, reverse orange to red.....*P. aureo-flavum* Bi-  
 ourge, 42.  
 XXIV. Conidia globose.....XXV.

- XXVa. Colonies fairly broadly spreading, wrinkled, with bristly prostrate or ascending ropes or almost coremia, pale green or gray green; conidia 2 to  $2.5\mu$ .....*P. Niklewskii* Zal. 45.
- XXVb. Colonies spreading broadly, with a felt of hyphae and ropes of hyphae, with alternating zones of pale green conidial areas and colorless sterile areas, conidia 2 to  $2.5\mu$ .....*P. Adametzi* Zal. 46.
- XXVc. Colonies forming felts up to 3 to 4 mm. deep, composed of fine hyphae, with outer margin white, and central area vinaceous with vinaceous drops, reverse vinaceous, conidiophores short, sometimes branched, conidia 2, 2.5 or even  $3\mu$ .....*P. vinaceum* Gilman and Abbott, 47.
- XXVd. Colonies forming thin tough felts, gray green, bluish green or gray; reverse deep orange; conidia 2.5 to  $3\mu$ .....*C. griseus* Sopp, 48.
- XXVe. Colonies erumpent upon leaves of cane.....*P. platense* Speg., 49.
- XXVI. Colonies velvety in appearance but with gradation from trailing hyphae to trailing ropes of hyphae.....XXVII.
- XXVII. Conidia subglobose to globose, smooth.....XXVIII.
- XXVII. Conidia subglobose to globose, rough or granular.....XXIX.
- XXVIIIa. Colonies velvety white to drab, scarcely greenish; conidia 2.5 to  $3\mu$ .....*P. decumbens* Thom, 50.
- XXVIIIb. Colonies bluish to yellowish green to drab; reverse yellowish to greenish to deep blackish green; conidia about 2.5 to  $3\mu$  by up to  $3\mu$ .....*P. fellutanum* Biourge, 52.
- XXVIIIc. Colonies evanescently bluish green to grayish sulphur yellow; reverse citrine; conidia subglobose 2.8 by 2.2, or 2 to  $3\mu$ .....*P. citreo-sulfuratum* Biourge, 51.
- XXVIIIId. Colonies almost velvety, pale bluish green, to green, to brown; conidia about  $2.5\mu$  in diameter.....*P. citreo-viride* Biourge, 53.
- XXVIIIe. Colonies gray to very pale green; reverse to greenish shades; to purple violet; conidia subglobose 1.5 to  $3\mu$ .....*P. phaeo-janthinelum* Biourge, 54.

- XXVIII. Colonies producing floccose felts of delicate hyphae, gray with faintly green marginal zones; conidia elliptical to globose about  $2\mu$ .....*P. cinerascens* Biourge, 55.
- Not *P. cinerascens* Gilman and Abbott.
- XXIX. Conidia granular to echinulate.
- XXIXa. Colonies appearing velvety spreading, zonate, in bluish-green shades becoming yellowish gray in age; conidia about  $3\mu$  delicately punctate as seen under oil immersion.....*P. Paczoskii* Zal. 56.
- XXIXb. Colonies spreading broadly, colorless to pale flesh color, with some green conidial area in the central area; conidiophore walls pitted or rough; conidia  $2.5$  to  $3\mu$ , faintly roughened.....*P. Terlikowskii* Zal. 57.
- XXIXc. Colonies mostly rosy in color with conidia green only on bread and other special media; reverse in deep red shades, conidia elliptical  $3.5$  to  $5$  by  $2.4$  to  $4\mu$ , delicately roughened.....*P. roseo-cinnabarium* Biourge, 58.
- XXX. Colonies velvety or velutinous.....XXXI.
- XXXI. Colonies velvety to the eye or handlens but with trailing hyphae and more or less ropy as seen under the microscope.....XXVI to XXIX.
- XXXI. Colonies velvety, with simple conidiophores mainly rising from submerged hyphae.....XXXII.
- XXXII. Conidia globose or subglobose.....XL.
- XXXII. Conidia elliptical.....XXXIII.
- XXXIII. Conidia elliptical rough or spinulose.....XXXIV.
- XXXIII. Conidia elliptical smooth.....XXXV.
- XXXIV. Colonies deep blue green, reverse and agar yellow to tan; conidia  $3$  to  $4$  by  $2.6$  to  $3\mu$ , rough when ripe.....*P. lividum* Westling, 65.
- XXXIV. Colonies in dark gray to fuliginous shades; reverse yellow; penicillus part monoverticillate, mostly complex divaricate; conidia  $3$  to  $3.5$  by  $2.2$  to  $2.7\mu$  rough.....*P. nigricans* (echinatum Dale) Bainier, 245, XVII.

- XXXV. Colonies closely velvety 100 to 200 $\mu$  deep, in gray green shades.....XXXVI.
- XXXV. Colonies closely velvety 100 to 200 rarely 300 $\mu$  deep in blue green shades.....XXXVII.
- XXXVIa. Closely velvety; 100 to 200 $\mu$  deep. White or nearly so on *Hydrocharis morsus ranae*, stalk short; sterigmata divergent; conidia 3 to 3.5 $\mu$  in long axis in short divergent chains.....*P. morsus-ranae*  
Corda, 597, XXV.
- XXXVIb. Gray green; with reverse yellow to liver brown; conidia 3 to 4 by 2.5 to 3 $\mu$  in loosely parallel chains.....*P. dierckxii* Biourge, 66.
- XXXVIc. Gray green to green; wrinkled and buckled; reverse vinaceous fawn; conidia 3 to 4.5 (5) by 2 to 3 $\mu$  in loosely parallel chains.....*P. turbatum* Westling, 67.
- XXXVIId. Colonies gray-green to gray, with bluish tinge at the margin, in age dark or fuliginous gray; reverse yellow to reddish orange; conidia 2.2 to 3.8 by 1.8 to 3 $\mu$ .....*P. griseo-atrum* Biourge. See Chapter XXVI.
- XXXVIe. Colonies gray green with bluish margin, showing an occasional metula 25 to 30 by 2.4 $\mu$ , sterigmata 8 to 13 by 2 to 2.5 $\mu$ ; conidia in column 3 to 3.2 by 2 to 2.4 $\mu$ ; reverse ultimately reddish-violet.....*P. aurantio-violetum*, 68.
- XXXVII. Closely velvety, 100 to 200 rarely 300 $\mu$  deep, bluish green series.....XXXVIII.
- XXXVIIIa. Colonies showing restricted growth, dark green, with margin bluish green, later gray green; reverse yellow to janthino-violaceous, substratum yellow, stalks?; conidia ovate 2 to 3.5 by 1.6 to 3 $\mu$ , occasionally much larger, smooth.....*P. jantho-citrinum*  
Biourge, 69.
- XXXVIIIb. Colonies restricted in growth, gray green to ferruginous; reverse uncolored to reddish or brownish red, conidia 2.3 to 3.2 by 2.2 to 2.4 (or up to 2.8).....*P. subcinereum*  
Westling, 70.

- XXXVIIIc. Colonies bluish gray green, spreading in thin radiating lines at margin; conidia 1.5 to 3.5 by 1.5 to 3.....*P. citreo-nigrum* Dierckx, 71.
- XXXVIIIId. Colonies dark gray green, with margin blue green to orange brown in age; reverse yellow to reddish brown; conidiophores short from creeping hyphae often partly branched; conidia 3 to 3.2 $\mu$  by 2 to 2.4 $\mu$ .....*P. aurantio-violaceum* Biourge, 68.
- XXXVIIIe. Colonies velvety, dull bluish green, buckled and wrinkled, 100 to 200 or 300 $\mu$  deep, agar citrine (aspect of *P. citrinum* CT.); overgrowth of long hyphae, occasional branch, cited but not figured by Biourge; conidia (2) 3 to 4 $\mu$  in long axis or 2 to 3.5 by 1.8 to 2.8 $\mu$ .....*P. implicatum* Biourge, 72.
- Colonies as above except with margin yellow to orange.....*P. implicatum* var. *aureomarginatum* n. var. Thom, 73.
- XL. Conidia globose or subglobose.....XLI.
- XLI. Reverse of colony quickly and intensely red..XLII.
- XLII. Reverse of colony not quickly and intensely red.....XLIV.
- XLIIa. Reverse red; conidia 4 $\mu$  rough forming dense crusts.....*P. favedkamp* Kap Laboratory?, 75
- XLIIb. Reverse rose; conidia 3 to 3.5 $\mu$ .....*P. Bruntzii* Sartory. See 33.
- XLIIc. Reverse red; drops yellow to brick red; conidia 2.2 to 2.5 $\mu$ .....*P. multicolor* G. M. and P., 75.
- XLIIId. Colonies blue then green, to brown; with red spots in reverse; conidia 2 to 3.5 $\mu$ .....*P. rubro-punctatum* Dierckx, 610, XXV.
- XLIIe. Colonies closely velvety, glaucous gray with white margin and closely felted mycelium; reverse and agar orange to deep mahogany red; conidia globose 2 to 2.5 $\mu$ —from the Bainier collection.....*C. cesiae* B. and S., 19.
- XLIV. Conidiophore walls rough.....XLV.
- XLIV. Conidiophore walls smooth.....L.

- XLV. Colonies some shade of green or blue green....XLVI.
- XLVIa. Colonies blue-green to dark green to brown in age, velvety, wrinkled; reverse yellow brown; bad odor on all substrata, conidiophores rough, coarse; conidia  $4\mu$ .....*C. foetens* Sopp, 77.
- XLVIb. Colonies velvety, bluish green, then gray green, about  $300\mu$  deep, with reverse yellow to reddish brown in spots; conidiophores and sterigmata rough (squamulose Biourge); conidia 2.5 to  $4.4\mu$ .....*P. flavi-dorsum* Biourge, 92.
- XLVIc. Colonies green to avellaneous in age, densely velvety, orange, then deep brown below, occurs as brown areas on Swiss cheese; conidiophores about  $100\mu$  long with walls rough; conidia  $3.5\mu$  (C.T.).— $4\mu$  (Staub.)....*P. casei* Staub. See 155, XIV.
- XLVID. Synonym.....*P. biourgei* Dierckx, 156, XIV.
- L. Conidia globose; conidiophore walls smooth..LI.
- LI. Conidial chains in compact columns.....LII.
- LI. Conidial chains loosely parallel.....See 25 et seq.
- LII. Colonies restrictedly or slowly growing.....LIII.
- LII. Colonies broadly spreading.....LIV.
- LIII. Reverse uncolored.
- LIIIa. Colonies slowly forming a thin colorless mycelium with few and scattered columnar penicilli.....*P. columnare* n. sp., 78.
- LIIIb. Colonies pale gray to olive green.....4967.7 Melin. var of *P. spinulosum*, 25.
- LIIIc. Reverse yellow.....5001.13b.
- LIV. Colonies spreading, velvety, conidial chains forming columns.....LV.
- LV. Conidia smooth.....LVI.
- LV. Conidia rough or spinulose.....LIX.
- LVI. Colonies brown, or avellaneous, without green color.....*P. silvaticum* Oudemans, 611, XXV.,
- LVI. Colonies blue green to green to brown; reverse yellow to orange or reddish orange; conidia 2.6 to  $3\mu$ .....*P. glabrum* series, 80, I VII.



- LVII. Related to *P. frequentans*; conidia reported as smooth.....LVIII.
- LVII. Related to *P. frequentans*; conidia reported as rough.....LIX.
- LVIIIa. Colonies blue gray; reverse uncolored; conidia 3 to 4 $\mu$ .....*C. albicans* Sopp, 81.
- LVIIIb. Colonies 100 to 200 $\mu$  deep blue green to olive green; reverse orange brown; conidia about 3 $\mu$ .....*P. aurantio-brunneum* Dierckx, 82.
- LVIIIc. Colonies 400 to 600 $\mu$  deep, dull green; reverse yellowish toward brown; conidia 3 to 4 $\mu$ ....*P. candido-fulvum* Dierckx, 83.
- LVIIId. Colonies 200 to 300 $\mu$  deep, blue green to brown; reverse yellow to orange brown; conidia 2 to 3 $\mu$ .....*P. citrinum* fide Biourge, 84.
- LVIIIe. Colonies zonate, gray green; conidia 3 to 4 $\mu$ ....*P. geophilum* Oudemans, 85.
- LVIII f. Colonies green; reverse in brown shades; conidia 2.5 to 3 $\mu$ .....*P. glabrum* (Wehmer?) Westling, 86.
- LVIIIg. Colonies deep dark green to gray, becoming hemizonate; reverse in orange shades; conidia about 2.5 to 3 $\mu$ .....*P. Oledzkii* Zal, 87.
- LVIIIh. Colonies bristly gray green to gray; reverse yellow; conidia 3 $\mu$ .....*C. robustus* Sopp, 88.
- LVIIIi. Colonies gray green, about 300 $\mu$  deep; reverse yellowish brown to brick red; conidia ovoid to subglobose 2.8 to 3.2 $\mu$  in long axis.....*P. sublateralitium* Biourge, 89.
- LVIIIj. Colonies in green shades becoming orange brown shades in age; reverse in orange yellow; conidia 2.5 to 3 $\mu$ .....*P. Szulczewskii* Zal, 90.
- LIX. Conidia reported as rough.....
- LIXa. Colonies in blue green to brown shades; reverse yellow to brown; conidia 2.5 to 4.8 $\mu$  rough.....*P. baiiolum* Biourge, 91.
- LIXb. Colonies in blue green to gray green shades; reverse colorless or with spots of yellow to reddish brown; conidia 3 to 3.5 faintly echinulate.....*P. flavi-dorsum* Biourge, 92.

Section I. Stricta: Conidiophore mostly unbranched; branching when present irregular, occurring only on a small part of the conidiophores and lacking the suggestion of a terminal verticil.

Subsection I. Stricta-Sclerotigena: Sclerotia or undeveloped perithecia have been described in a small number of species which are arbitrarily brought together here since true affinities elsewhere have not been established.

1. *P. thomii* Maire, Bul. Soc. Hist. Nat. Afrique du Nord. 8: pp. 189-192. 1917. Compare no. 29 in Thom, C., U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118:78. 1910. Not *P. thomii* Zaleski q.v. (Compare our fig. 19.)

Maire's Latin diagnosis, p. 192, may be translated: Colonies in bean agar with cane sugar, sordid (amoene) gray green, quickly and broadly

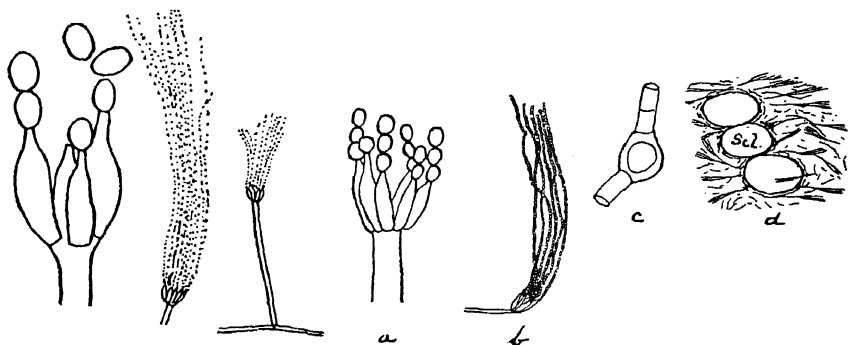


FIG. 19. *P. thomii* Maire: a, the verticil of sterigmata; b, the penicillus with parallel chains of conidia; c, germinating conidium with two germ tubes; d, diagrammatic sketch of part of colony showing three sclerotia, *Scl*, with hyphae and conidiophores and penicilli in the field.

spreading in the substratum with growing margin broad white; aerial growth composed of crowded conidiophores; reverse of colony white then salmon; conidiophores 40 to 300 by 1.5 to 2.5 $\mu$  unbranched, dilated to 4 $\mu$  at the apex and bearing a verticil of 4 to 8 sterigmata 7 to 9 by 1.5 to 2 $\mu$  subfusiform; conidia in chains loosely columnar in arrangement, 3 to 3.5 by 2 to 2.5 $\mu$ , very pale greenish, thin walled, smooth; sclerotia salmon, subglobose to ellipsoid, 100 to 250 by 100 to 200 $\mu$ , frequently confluent; gelatine not liquefied; cultures acid to litmus.

Species found upon an *Amanita ovoidea* in Algeria.

Maire notes slight divergences from Thom's description of his no. 29 in which gelatine was reported as liquefied, sterigmata more numerous,

conidiophores coarser, and the reverse of colonies yellow but regards these as minor or racial characters not warranting separation.

While Thom's original no. 29 has been lost, several races or strains with this morphology have been collected and when compared showed only minor cultural differences as suggested by Maire. While growing readily in culture if cared for and regularly transferred, all of them have been lost, showing that the species requires more care than many other forms which have been kept for many years.

Cultures received from Professor Atkinson (Ithaca, N. Y.), no. 22605, 22572, and 22791, appeared to be identical with no. 30. Cultures of nos. 29 and 30, together leave so small differences as to lead to the conclusion that they are only races or strains of a single species. This organism (no. 29) has also been cultivated by Dr. M. T. Cook who found that the sclerotia would germinate in the spring after exposure through the winter. In no case, has any development into a perithecium been seen.

Another culture was received from Dr. C. E. Lewis at Orono, Maine, which gave the same conidial morphology. This organism produced sclerotia abundantly at first, but gradually lost the power to do so. The same observation holds for the form discussed in a previous paper as no. 30. From cultures apparently pure at first, two strains were gradually separated. One of these corresponded closely with the one listed as no. 29 which continued to produce sclerotia. The other became eventually entirely conidial in form. Both of these retained the same morphology of the conidial apparatus. None of the experiments performed indicated any evidence of sexuality in these strains. Another culture was made from a moldy chestnut bur collected by Dr. A. F. Blakeslee at Cold Spring Harbor, New York. This proved to be a mixture of the sclerotium form described as no. 29 (B. A. I. Bul. 118) and an organism received from Amsterdam as *Citromyces glaber*, but no relation between the two forms was established. The same sclerotium producing species was also sent from England by Miss Dale, (1912) and reported by her (D2 p. 51).

The original culture described by Thom as no. 29 without name was isolated from a rotting mushroom at Storrs, Connecticut; the culture was finally lost. Maire described his culture from Algeria giving the specific name *P. thomii*.

More recently in a series of cultures contributed by Dr. Povah from Evanston, Illinois (5001.3c, 5a and 19b) pink sclerotia appeared in no. 3c upon certain media together with more abundant conidiophores than

*P. thomii*; in no. 19b the sclerotia were paler in color and the conidia much more abundant; in no. 5a, conidia were very abundant but no sclerotia were found. These cultures without sclerotia satisfy the description of Biourge's *P. aurantio-violaceum* fairly well.

Further study of this series of sclerotium producers will be necessary before the true lines of separation can be found or they can be put into their proper relationship with some group of ascomycetes.

3. *Citromyces tuberifer* Rostrup. Bot. Tids. 29, p. 32-41, fig. 1908.

Syn.? *C. tubifer* Rostrup 1908 in Biourge's List.

The Latin diagnosis p. 39 may be translated: Colonies at first glaucous, then olivaceous, with sterile hyphae creeping, septate branched; conidiophores unbranched, erect, septate, 115 to 170 by 3 to 5 $\mu$ , with apex dilated to a clavate vesicle 6 to 9 $\mu$  in diameter, bearing up to 10 sterigmata at the tip 6 to 8 by 2 $\mu$ ; conidia almost globose, hyaline in long chains 3 $\mu$  in diameter; sclerotia very abundant, globose or ovate, or more or less irregular pale—flesh colored (*incarnatis*), 180 to 660 more commonly about 400 $\mu$  in diameter.

This is cited by Klöcker, A. Garungsorganismen 3 aufl. 311, as a sclerotium forming "*Citromyces*" discovered by Klöcker himself, later also by Rostrup who described it. *C. tubifer* in Biourge is probably a misspelling.

*The following species described by Sopp as sclerotium producers have never been identified by others. They fall arbitrarily in this place.*

4. *C. albo-roseum* Sopp. Monogr. pp. 122-125. Taf. XV, fig. 106 and Taf. XXII, fig. 7. 1912.

Colonies on meat-peptone-sugar-gelatine, with tough, wrinkled mycelium white above and in reverse slowly developing abundant red "perithecia" (= sclerotia) in the center with numerous colorless drops of fluid, then clear green conidial areas develop upon the mycelium outside the perithecial areas, giving the effect of patches of red mixed with patches of green; hyphae coarse, septate; reverse pale reddish; conidiophore unbranched, septate, with moderate vesicular apex; sterigmata described as flaskshaped; conidia 5 to 7 $\mu$  described as small and round, (figures in relation to other parts as in other species leading to the suggestion of error in the measurements reported—C.T.); sclerotia (called perithecia by Sopp) prominent, giving red color to the colony, dense, hard, pseudoparenchymatous, consisting of an outer mass of thick-walled cells, and inner more hyphal mass but no asci found.

Species found in earth in west Norway; colonies grew between 1° and 40°C. with optimum of 15 to 20°C., grew well in meat-peptone gelatine without sugar, in milk, broth, with and without tartaric acid, in wort, on potato, on rice and on bread producing little odor or taste and color in the substratum.

5. *C. coeruleus* Sopp. Monogr. pp. 110–112. Taf. XIII, fig. 95 and XXII, fig. 1. 1912.

Colonies at first steel-blue, later gray green and finally brown, closely velvety, with center depressed and more or less radiately wrinkled ("gray wrinkles"), and with thin white fibrous margin; exuding very small hyaline drops; odor offensive, suggestive of cat's urine; mycelium forming a thin felt, becoming chrome to cadmium yellow in reverse; conidiophores usually *Citromyces*-like with apex much enlarged, occasionally branched from the second node producing one or two secondary beads or single sterigmata, and more rarely producing one or more secondary conidiophores; sterigmata 5 to 25 $\mu$  in number, rather large diameter; conidia about 5 $\mu$  in diameter; sclerotia formed only on colostrum.

Species was found in earth, in Norway. It grew from +1° to +40°C., liquefied gelatine, produced good growth on milk, meat-peptone broth, potato, rice, bread, and tannin solution, and usually produced an offensive odor.

*C. coeruleum* Bainier and Sartory is given by error in Biourge's "List onomastique" Monogr., p. 102.

*Sub-section 2. Stricta-floccosa: Colonies not velvety in appearance, but with a floccose, cottony mass of aerial hyphae bearing conidiophores mostly as branches rather than by origin from submerged hyphae. Section leads to the soil penicillia and to the Lanata in the Asymmetrica.*

\* Colonies in gray not in green shades.

6. *P. restrictum* Gilman and Abbott. Iowa State College Jour. Sci. 1: 297, fig. 32. 1927. See our figure 20.

Colonies on Czapek's solution agar, forming gray or neutral gray to almost black closely woven felts, crenulate and lighter at the margin, restricted in growth showing no green color, consisting of fine hyphae, tearing easily, showing abundant crystal drops at the line between the deeper central area and the light or white marginal zone; reverse colorless; conidiophores borne as branches of aerial loops or trailing hyphae,

commonly very short, often about  $10\mu$ , rarely  $100\mu$  long, penicilli consisting of 1 verticil of sterigmata bearing conidial chains loosely parallel to divergent, or occasionally in a fairly close column up to 50 to  $75\mu$  long; sterigmata 5 to  $6\mu$  long, tapering to sharp points; conidia 2 to 2.5 or up to  $3\mu$  in diameter, globose, delicately echinulate, and often showing a connective.

Type received from Gilman, our no. 4894.14, reported as found by him in Louisiana soil. The above diagnosis is redrawn from our own cultures but varies only slightly from Abbott's description.

5048.18a from Texas soil reproduced this species in general but was lighter gray in colony color and produced pale cinnamon to buff color in the outer areas in reverse.

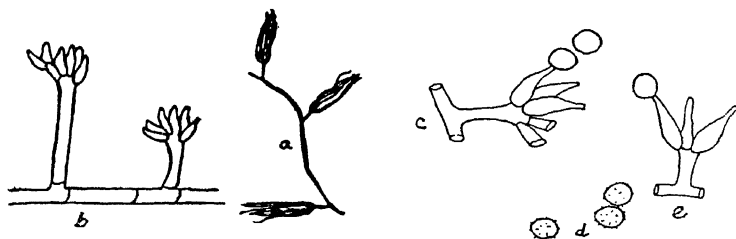


FIG. 20. *P. restrictum* Gilman and Abbott: a and b, Gilman and Abbott's detail and habit figures; c, e, penicilli; d spiny conidia.

Another species with this morphology was isolated from a peat bog by Dr. C. L. Shear—no. 3553:

Colonies on Czapek's solution agar azonate floccose forming a felt up to 1 mm. or more in thickness in central areas, thinning out at the broad white margin, with conidial areas at first pale greenish gray Ridgway LII. 35'''' Hathi gray to stone gray passing into neutral gray, or olive gray shades varying in different cultures; aerial hyphae about  $2\mu$  in diameter, with granular walls; reverse pale citrine, citrine to clay yellows; drops large, abundant, colorless, formed in the conidial areas and leaving holes or pits in the surface growth after drying; odor evident but weak, aromatic; conidiophores mostly very short 10 to 20 more rarely up to  $50\mu$  by about  $2\mu$  with walls granular; penicilli mostly monoverticillate with 1 or 2 secondary penicilli on unequal branches upon some conidiophores in every field; sterigmata 6 to  $8\mu$  long, sharp pointed, few in the verticil; conidia rough or spinulose 2, 2.5 to  $3\mu$  or in old cultures  $3.5\mu$ , in parallel, diverging or in age tangled chains.

*\*\*Colonies showing some shades of green: a. Conidia more than  $4\mu$  in long axis; 1. Reverse or substratum red or reddish.*

10. *C. purpurrescens* Sopp. Monogr. pp. 117-119. Taf. XIV, fig. 102; Taf. XXII, fig. 4. 1912.

Colonies on meat-peptone-sugar-gelatine, with thick mass of mycelium somewhat folded or wrinkled, wooly or floccose, white, studded with dark red drops, and producing dark olive-green conidial areas as spots or marginal areas; reverse at first somewhat reddish, then purple, finally black, with color production reduced or delayed in some cultures; hyphae commonly fine; conidiophores moderately coarse, septate, mostly producing 1-verticil of sterigmata, occasionally with 1 to several branches at the uppermost node; sterigmata flaskshaped, in great numbers in the verticil, occasionally bearing secondary sterigmata; conidia globose, echinulate, about  $6\mu$  in diameter, produced in great abundance and from the figure apparently forming columns; perithecia and sclerotia not found.

Species found in earth in Norway; colonies grew from  $2^{\circ}$  to  $35^{\circ}\text{C}.$ , not at  $37^{\circ}\text{C}.$ , with the optimum between  $20^{\circ}$  and  $25^{\circ}\text{C}.$ , grew well in Koch's gelatine with and without sugar, also in agar, milk, wort, and bread, but poorly on potato and rice and sawdust; known only in Sopp's description.

11. *C. rubescens* Sopp. Monogr. pp. 126-128. Taf. XIII, fig. 97; Taf. XXII, fig. 9. 1912.

Colonies on meat-peptone-sugar-gelatine, at first blue-green, then olive green and finally brown, appearing as colored patches on islands on a white buckled or folded mycelium floating on liquefied gelatine; mycelium tough, felted or leathery, in reverse bluish then chamois color in old colonies; gelatine liquefied and colored wine red; potato colored reddish-yellow and rice greenish yellow; conidiophores moderately coarse figured as short branches of creeping ascending or trailing aerial hyphae, with moderately vesiculose apex; sterigmata in a single verticil, comparatively long and coarse, 5 to 10 in the verticil; conidia brownish gray, globose, given in table, p. 109, as 5 by  $4\mu$  (figures not repeated in the text); perithecia not found.

Species found in cellar-earth in east Norway; colonies grew at  $1^{\circ}\text{C}.$  and ceased to grow at  $30^{\circ}\text{C}.$ , with optimum at "room" temperature, grew well on potato and rice, but poorly upon most of the media tested; known only in Sopp's description.

12. *C. virido-albus* Sopp. Monogr. pp. 131-132. Taf. XIII, fig. 98; Taf. XXII, fig. 12. 1912.

Colonies on meat-peptone-sugar-gelatine, with blue-green to olive green velvety to somewhat fibrous or mealy conidial areas upon mycelium wrinkled, tough, almost leathery, in reverse reddish to chamois-red, and finally brown; gelatine liquefied and wine red; conidiophores short, crowded (?), slender, occasionally branched (but not so figured), with only slightly vesicular apex; sterigmata largest at the upper end (not so figured—C.T.) 5 to 10 in the verticil, short; conidia large, somewhat irregular, brownish under the microscope, given as 3 to  $4\mu$  in the description and as 3 to  $7\mu$  in the table, p. 109; perithecia not found.

Species found in earth in west Norway. Colonies grew fairly well at  $1^{\circ}\text{C}.$ , but best at "room" temperature, grew well in gelatine and agar, in milk in which the red color of the mycelium was more prominent, upon potato with yellow green conidial areas and golden drops of fluid and upon rice with mycelium at first yellowish then reddish; conidia survived in the laboratory more than three years; species known only in Sopp's description.

In description conidia are given as 3 to  $4\mu$ , but in the table, p. 109, as 3 to  $7\mu$ , the descriptive term "grosz" on p. 131 and 132, would hardly apply to conidia 3 to  $4\mu$  hence this is probably a misprint.

2. *Reverse or substratum not red.*

15. *C. olivaceus* Sopp. Monogr. pp. 129-131. Taf. XIV, fig. 99; Taf. XXII, fig. 11. 1912.

Colonies on meat-peptone-sugar-gelatine olive green, velvety, showing abundant crystal drops of fluid during the growing period and producing great masses of conidia; mycelium firm, woody, wrinkled close felted, gray-white, submerged (?), in reverse yellowish to greenish white; gelatine liquefied; odor indefinite; conidiophores long, rather slender, few septate, with moderately vesicular apex; sterigmata pointed at both ends, comparatively large! conidia globose, smooth, almost black, 6 by  $5\mu$  (in table, p. 109, no measurements given in text), produced in great abundance, and from the figures probably in columns; perithecia not found.

Species found upon leather upon a manure pile; growth was abundant at  $1^{\circ}\text{C}.$ , but without conidial formation; "room" temperature appeared to be optimum. Colonies grew well upon gelatine and agar, in milk,



in wort, upon potato, upon rice, in tannin solution and in oak sawdust. Conidia remained viable in the laboratory about three years. The species is known only in Sopp's description.

16. *C. fuscus* Sopp. Monogr. pp. 120-122. Taf. XIV, fig. 100; Taf. XXII, fig. 6. 1912.

Colonies upon meat-peptone-sugar-gelatine with mycelium spreading rapidly over the substratum, forming a thin close-felted but tough wrinkled mass, quickly becoming dark olive green with the development of the conidial area which becomes dark or fuscous in age; reverse at first reddish yellow (chamois) later with greenish shades ultimately almost black; gelatine colored reddish brown; odor little or none; conidiophores arising from prostrate coarse hyphae, at the surface of the mycelium, septate, fairly slender, and moderately long, with small vesicular enlargement at the apex bearing 1 to 15 sterigmata; sterigmata borne all over the vesicular area as in *Aspergillus*, and with their chains of conidia divergent, occasionally branched and bearing secondary sterigmata; conidia showing a definite connective, echinulate, globose, brown, in age smoother than when young (!—C.T.), 5 to 6 $\mu$  in diameter; perithecia not found.

Species found in earth in west Norway. Colonies grew from 2° to 40°C., produced good colonies in milk, urine and wort, but poorly on potato, rice and bread. The species is known only in Sopp's description.

*Conidia less than 4.5 $\mu$  in long axis. 1. Reverse or substratum or both red or reddish.*

17. *C. sanguifluus* Sopp. Monogr. pp. 115-117. Taf. XV, fig. 105; Taf. XXII, fig. 3. 1912.

Colonies on meat-peptone-sugar-gelatine, with tough leathery mycelium irregularly wrinkled and folded, at first more or less yellow, later changing to reddish then red, producing loosely velvety, pale greenish conidial areas, and exuding abundant blood red drops of fluid; reverse and gelatine at first yellowish red, then progressively deeper red until almost black with coloring matter difficult to remove from cloth; hyphae comparatively fine; conidiophores slender, uniform, such branched with branches short, diverging and bearing single verticils of sterigmata, usually curved, and few in the verticil; conidia globose 1.5 $\mu$  (in diagnostic description, given as 3 $\mu$  in Sopp's "Key") in divergent chains; perithecia and sclerotia not found.

Species found in earth in east Norway; colonies grew from  $+8^{\circ}$  to  $40^{\circ}\text{C}$ . with optimum between  $25$  and  $30^{\circ}$ , grew well in urine, in meat-peptone broth, upon potato and upon bread. Upon tannin solution, colonies are small, smooth, green to dirty green without any trace of red color; spores remained viable in the laboratory about three years. This species is recorded as difficult to cultivate and has not been reported by others although Biourge makes it a synonym of *P. roseo-purpureum* Dierckx.

If Biourge is correct in identifying this form with Dierckx's *P. roseo-purpureum*, Sopp's name should be retained and *C. cesiae* and *P. roseo-purpureum* Dierckx replaced by *P. sanguifluus* (Sopp) Biourge.

18. *P. roseo-purpureum* Dierckx. Soc. Scientifique Bruxelles 25: p. 86. 1901.

In Biourge Monogr. La Cellule 33: fasc. 1. pp. 317-319; Col. Pl. X. Cart. 8; Pl. XVI. fig. 96. 1923; *ibid.* 36: 482, 1925.

Synonymy according to Biourge: *Citromyces cesiae* Bainier; *Citromyces sanguifluus* Sopp.

Colonies on wort gelatine tomentose to lanose, elaborately wrinkled; green, with more or less yellow, soon beautifully variegated sulphur yellow, to rosy or flesh color, with drops carmine to deep blood red; coremia none; reverse at first orange yellow, then reddish brown, blood red, to dark purple red; odor none; conidiophores 10 to 30 less commonly 50 by  $2.4$  to  $2.8\mu$ , with all walls smooth, with apex scarcely enlarged, arising from decumbent hyphae; sterigmata 6 to 13 by  $2.2$  to  $2.8\mu$ , in verticils of 4 to 12; conidia subglobose 2 to  $3\mu$ , rarely  $3.5$  or even  $4\mu$ .

Biourge no. 8 (our no. 4733.108) grown upon Czapek's solution agar produced colonies pinkish buff in color, with closely felted mycelium 300 to  $400\mu$  thick consisting of fine hyphae less than  $2\mu$  in diameter, wrinkled or buckled, rising fairly abruptly from the substratum, in age becoming deeper (up to  $600\mu$  or even more) in center and tufted almost Isaria-like; drops abundant in pink to red colors; reverse yellow to vinaceous to deep red, with agar paler and deeply penetrated by mycelium; conidiophores arising as very short branches from aerial hyphae in the close woven felt, less than 100 by about  $2\mu$ , or occasionally rising from submerged hyphae and up to  $100\mu$  long; sterigmata 6 to 8 occasionally to  $12\mu$  in verticils of 5 to 8; conidia about  $2\mu$  in diameter.

Biourge makes this synonymous with *C. sanguifluus* Sopp and *C. cesiae* Bainier in which case Sopp's species being well described in 1912 should carry the name.

19. *C. cesiae* Bainier and Sartory. Bul. Soc. Mycol. France 29: p. 148-154. P. V. fig. 4-6. 1913.

Colonies upon licorice sticks floccose, in a comparative thick mass spreading rapidly with conidial areas in blue or blue green to dark grayish blue, quickly producing drops of transpired fluid rose, or red orange or red on potato cultures; reverse and substratum such as agar or potato, orange rose, shades of reddish or red; conidiophores produced as branches or as terminal segment of creeping or ascending (not erect) hyphae, about  $2.8\mu$  in diameter, and uneven in length, only slightly enlarged at the apex, and bearing verticils of 6 to 12 sterigmata about  $8\mu$  in length; conidia globose about  $2\mu$ , in loosely parallel to divergent chains (fig. 28c).

Species found in the tunnels made by *Cesia apiformis* in a poplar tree. Cultures grew best at  $24^{\circ}$  to  $25^{\circ}\text{C}.$ , grew well on all the usual media with the production of red coloring material which is soluble in fat-solvents suggesting the lipochromes of Zopf. Colonies coagulated milk, liquefied gelatine, did not act upon starch and produced citric acid from glucose.

Culture 4640.422, labeled *C. cesiae* as received in our set of the Bainier collection proved to be *C. cyaneus*. If Biourge is right in regarding the species as the same as Dierckx's *P. roseo-purpureum*, both names should be replaced by Sopp's *C. sanguifluus*.

*Reverse not in red or reddish colors.*

A considerable group of strains has been seen with the presence of a loose aerial basal network becoming fairly heavy in some, reduced to negligible in others but accompanied by the production of monoverticillate penicilli with chains of conidia in a parallel or loosely columnar arrangement together with a common aspect or cultural habit. Several described species belong in the group differing in details of colony habit, color and in the presence or absence of spinulosity in the conidial wall. Thom's *P. spinulosum* may be taken as a type of the spinulose sub-series and Wehmer's *P. Pfefferianum* for the sub-series with smooth conidia. We doubt if there is any really sharp line between them.

#### 24. *P. Pfefferianum* series.

Conidia 2.5, 3 up to  $4\mu$  spinulose (often appearing smooth with low magnifications); reverse colorless or showing pale yellowish or even reddish tones.

A closely related series of races or strains differing in minor details, hence apparently described under several names is typified by *P. spinulosum* Thom. It was originally obtained in Wehmer's laboratory and has been frequently received as *Citromyces pfefferianus* Wehmer, but positive identity with Wehmer's species has not been established, although Pollacci went farther and reduced the 27 forms studied by him (apparently only one of them in culture) to the rank of synonyms or cultural varieties of Wehmer's species.

Wehmer in his recent letter to Dr. Raistrick refers to one of Dr. Raistrick's strains; "No. 68 hat noch am meisten Ähnlichkeit mit dem *C. Pfefferianus*." As a courtesy to Dr. Wehmer and believing that some one of this series of exceedingly variant organisms was described by him as *C. Pfefferianus* we are designating the series here by that name although it has stood in our notes for many years as the *P. spinulosum* series.

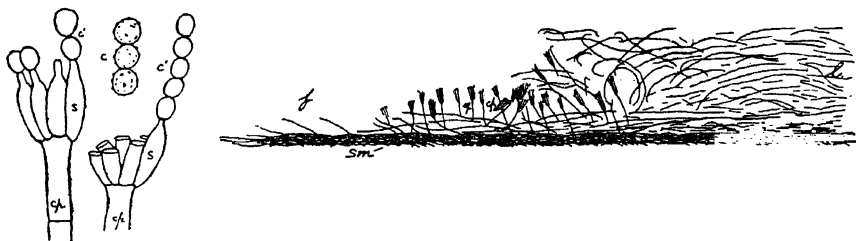


FIG. 21. *P. spinulosum* Thom; cp, conidiophore; s, verticils of sterigmata; c, spinulose conidia; c', young conidia smooth; at right, diagrammatic radial section of marginal area of colony, f, white margin; cp, conidiophores and penicilli with chains parallel or almost in columns; l, loose floccose area of mycelium.

*Conidia delicately spinulose.*

25. *P. spinulosum* Thom, in U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, p. 76, fig. 32. 1910. See figure 21.

Colonies upon gelatin, bean agar or Czapek's solution agar deep green, spreading broadly in the substratum with broad sterile margin when young; aerial portion consisting of conidiophores and sparsely floccose aerial hyphae; reverse of colony not discolored or at times showing a pinkish tinge; conidiophores 150–300 $\mu$  or longer by 3 to 3.5 $\mu$ , with apex enlarged to 5 $\mu$  in diameter, bearing a single verticil of sterigmata 9.5 to 11 by 2 to 3 $\mu$ ; penicillus a loose column of conidial chains up

to 300 or even 500 $\mu$  in length by 15 to 30 $\mu$ ; conidia pyriform to globose, 3.2 to 3.5 by 3.6 to 4 $\mu$ ; very thin walled, smooth at first then delicately spinulose or verrucose, yellowish green then almost smoky; liquefying gelatin slowly, with strongly acid reaction.

Found as a contamination of another species of *Penicillium* obtained in Doctor Wehmer's Laboratory at Hanover, Germany; easily recognized and cultivated. Although confirmation has not been possible there appears to be some evidence that this species is near if not identical the *Citromyces Pfefferianus* Wehmer Beitr. Kennt. Einh. Pilze I. 21, 1893.

Cultures giving the morphology and reactions of *P. spinulosum* have been many times isolated and studied by us. Others have been received from widely separated laboratories; these include soil cultures by Waksman (4078 series) at New Brunswick, New Jersey; one from Shear (4285.5) from cranberries; one from Linder (4754C75) among fungi from British Guiana; one from Putterill (4658.36.5) from South Africa; no. 7251.8.1 from wood at Forest Products Laboratory, Madison, Wisconsin; no. 4695 from Prof. C. M. Derick at McGill University, Montreal. A strain was isolated from moldy leaves collected for us by C. J. Koning of Bussum, Holland (2746.1a) from Spanderswoud. It produced a reddish reverse color in bean agar but was otherwise typical.

The following species more or less fully described probably belong in a series with *P. spinulosum*. Strain differences are often evident but as far as known to us the gradations occurring bridge all gaps too completely to leave much hope for separation on the basis of a morphological description.

26. *P. flavo-cinereum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 293-295; Col. Pl. VIII, Cart. 119; Pl. XIII, fig. 76; 1923.

In series with *P. spinulosum* Thom.

Biourge's diagnosis: Colonies on wort gelatine wrinkled, dark bluish green, soon (cinereous) gray, then dark fuscous; coremia none; reverse evanescently gray, sordid yellow, at times reddish; odor none; conidiophores with walls granular, 2 to 2.5 $\mu$  in diameter, penicillus 15 $\mu$  long or up to 40 to 60 $\mu$  long when branched, mostly in the simple form; metulae 20 to 35 $\mu$  long, commonly single, rarely in pairs, in age roughish granulate; sterigmata 8 to 11 by 3 to 4 $\mu$ , in groups of 4 to 8; conidia at first elliptical, 2.5 to 3.4 $\mu$  by 2 to 2.5 $\mu$ , then globose 3 to 4.5 $\mu$ , with walls granular;

Biourge's type no. 119 (our no. 4733.61) when grown upon Czapek's

solution agar produced colonies spreading broadly, covering the whole surface in the petri dish and even following up the sides and inside of the cover, forming a loosely floccose network of varying density, zonate in the outer areas, in color green, to gray and fuscous in age, with reverse uncolored; with microscopic details approximating those given by Biourge.

Our no. 2746.1a isolated from leaf mold collected by C. J. Koning for us in the forest known as "Spanderswoud" near Bussum, Holland, seems to have been close to Biourge's *P. flavo-cinereum*.

27. *P. fluitans* Tiegs. Ber. deut. bot. Gesellsch. 37: 499-501. 1919.

Colonies reported as growing on wort, bluish, blue green, to chocolate brown in age, with margin white to salmon or strawberry color, 5 mm. broad; velvety, plane; with the vegetative appearance of other *Penicillia*; reverse whitish or uncolored; hyphae 2 to 5 $\mu$  in diameter; conidiophores unbranched, 3 $\mu$  in diameter; sterigmata in one verticil, 7.5 to 10 by 2.7 to 3.4 $\mu$ ; flask-shaped, incurving; conidia globose, 2.6 to 3.5 $\mu$  with walls more or less roughened as seen under the oil immersion objective.

The species was found in the waste-water of a munitions factory in Germany and observed over several years growing as large masses of vegetative mycelium submerged in running water acidified with nitric acid; in experiments abundant vegetative mycelium developed with apparent inhibition of conidium formation in solutions containing 1.5 per cent (0.25N) nitric acid; conidium formation appeared only slowly and weakly in acid solutions.

This species was observed to occur in pure culture in one locality for several years as submerged tufts or skeins of vegetative mycelium attached to twigs and plants. The nitrogenous material and carbohydrates present in the water furnished nutrients apparently favorable for this species of *Penicillium*, since the *Penicillium* was replaced by other fungi when the flowing water became neutral.

28. *P. (Citromyces) Pfefferianum* (Wehmer) Westling. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 303-305; Col. Pl. VIII, Cart. 162; Pl. XII, fig. 72. 1923.

Biourge no. 162 (our no. 4733.95) when grown on Czapek's solution agar produced colonies loosely interwoven, lanose, spreading up to 500 $\mu$  deep, wrinkled and buckled, with white overgrowth, with growing margin broad, gray, dark gray green; reverse white to cream in petri dish cultures, tan or chamois in slant tubes with conidiophores up to

200 $\mu$  or more by 2 to 3.5 $\mu$  in diameter, swelling at apex to 5 to 7 $\mu$ ; sterigmata 11 to 12 $\mu$  long, some coarse and septate; conidia 3 to 4 $\mu$  delicate spinulose, with walls thin, in parallel, later tangled chains.

Culture 4733.95 is thus very close to *P. spinulosum*.

29. *P. roseo-maculatum* Biourge. Monogr. La Cellule 33; fasc 1, p. 301-303; Col. Pl. VIII, Cart. 43; Pl. XII, fig. 71. 1923.

Colonies on wort gelatine, wrinkled, more or less velvety in appearance, with margin broad, sublanose, white, at first bluish green (C.d.C. 417, 422), then in dark blue green to violaceous (indigo) shades, at length violaceous reddish brown; coremia none; reverse sordid greenish, then yellowish to fuscous; odor none; conidiophores 1.5 to 4 $\mu$  in diameter, mostly unbranched, or with a single branch low down on the stalk, arising from creeping hyphae; penicillus usually about 15 $\mu$  long, figured as a closely packed verticil of more or less unequal sterigmata; sterigmata 8 to 13 by 2.3 to 3.5 $\mu$ , in verticils of 5 to 12; conidia 2.5 to 4 by 2.4 to 3.5 $\mu$ ;

Biourge type no. 43 (our no. 4733.107) in our cultures grown on Czapek's solution agar proved to be *P. spinulosum* or near it as follows: Colonies deep lanose with considerable floccose; cottony, aerial mycelium and white overgrowth, in mass 400 to 600 $\mu$  deep; conidial areas dull green to dark sordid gray green; reverse and agar uncolored; conidiophores loosely ascending (not a velvety crowded area) bearing a simple verticil of sterigmata and chains of conidia closely arranged into a column 100 to 200 $\mu$  long; conidia with wall faintly spinulose about 3 $\mu$  or from 2.5 to 3.5 $\mu$ .

30. *P. viridi-dorsum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 306-307; Col. Pl. VIII, Cart. 183; Pl. XIII, fig. 75. 1923.

Colonies in wort gelatine, velvety much wrinkled, with margin of conidial area bluish green, then gray green within dark blue green almost black, finally dark brown, coremia none; reverse usually greenish verging toward reddish or hyacinth; odor none; conidiophores 2 to 4 $\mu$  in diameter, with apical cell granular, lower cell walls smooth, figured as 1 septate, lower half smooth, upper half granulate; sterigmata 10 to 13 by 3 $\mu$ , with walls granular, in verticils of five to eight; conidia globose, 3 to 4.5 $\mu$ , granular or echinulate;

Biourge's type no. 183 (our no. 4733.126) grown in Czapek's solution agar produced colonies up to 500 $\mu$  deep with conidiophores arising from loose fibrous web of hyphae creeping or partly submerged but

increasing with maturity of colony; reverse colorless; odor faint, fragrant; conidiophores 100 to 300 $\mu$  long ascending or tangled (not erect and parallel); conidial chains forming columns about 20 $\mu$  in diameter and 200 to 500 long, coiled and twisted not as parallel masses; conidia 2.5 to 3, possibly 3.5 $\mu$ , smooth or with double wall or traces of wrinkles or joints.

This is another variant of the *P. Pfefferianum* series.

*Conidia 2.5 to 4 $\mu$ , smooth, or nearly so or with markings not indicated in the description. P. Pfefferianum sub-series.*

31. *Citromyces Pfefferianus* Wehmer. Beitr. z. Kenntn. einheim. Pilze I: 22–24; Taf. I, figs. 1–13. 1893; Saccardo Sylloge Fungorum XI: 593; see also the descriptions which follow:

Wehmer's Latin diagnosis may be freely translated and supplemented from his German descriptive notes: sterile hyphae hyaline, septate effused and erect, branched; fertile hyphae simple or branched, scarcely or rarely septate, averaging 70 by 3 $\mu$  with apex more or less inflated to form a clavate vesicle 4 to 8 $\mu$  in diameter; sterigmata simple, hyaline 5 to 10 in number with apices acute, difficult to separate, 9 to 14 by 3 to 4 $\mu$ ; conidium bearing head penicillate; conidia globose, smooth in chains, abundant, greenish to gray 2.3 to 2.8 $\mu$  in diameter. Habitat: on rotting fruits of *Citrus medica*.

Verified cultures of this species have not been obtainable but several cultures from European workers indicate a belief that some strain near *P. spinulosum* was the basis of the description. Scrutiny of Wehmer's discussion gives few clues to absolute identification of the strain. In examining a large number of strains of this general group part of them appear to have smooth conidia hence to fit Wehmer's species. We have, therefore, separated the described species upon this character and present the whole group in the two sections.

32. *P. pfefferianum* (Wehmer) Pollacci. Atti Ist. Bot. Univ., Pavia ser. II, 16: 121–136, Pl. XVI. 1916. Syn. fide Pollacci *Citromyces pfefferianus*, Wehmer, *C. glaber* Wehmer, *C. sormanii* Carbone.

Sterile hyphae hyaline, septate, effused, erect, branched; conidiophores simple or branched, scarcely or rarely septate, 70 by 3 $\mu$  in diameter; sterigmata 9 to 14 by 2 to 4 $\mu$ , with apex acute, unbranched, 5 to 10 in verticil, separated with difficulty; conidia globose, 2.5 to 3 $\mu$  in diameter, in long chains in a solid column ("supra congregatis"), green then gray (cinereous).



Proveddi in Riv. di Biologia 8, fasc. 1, p. 17-18, 1926, amends Polacci's Latin description by giving the conidiophores as 70 to 130 $\mu$  long, the sterigmata as 3 to 12 in the verticil and the conidia as 2 to 3.5 $\mu$  in diameter.

Species producing citric acid by fermentation, are found in putrescent materials, in fruits, in sausage, in solutions of sugar, of citric acid, of oxalic acid, etc., in Europe.

33. *Citromyces bruntzii* Sartory. Compt. rend. soc. biol., Paris 76: 605-606. 1914.

On gelatine, milk and glucose containing media this species spreads slowly with a considerable fruiting mass in which it is very difficult to distinguish individual fruiting stalks. Conidiophores mono-verticillate with sterigmata, 10 to 12 in number, 9 to 10 $\mu$  in length; conidia in chains, globose, 3 to 3.5 $\mu$ . Gelatine was liquefied in twelve days, milk was coagulated with precipitation of casein and peptonization. Serum and albumen were not coagulated. Glucose was transformed into citric acid. This strain secretes a rose pigment with absorption bands near violet, for which the solubility is given.

Found on oranges from the Balearic Isles.

Cultures by Sartory.

This species has not been seen by us but the description would place it close to the *P. spinulosum* series from which it differs in lacking the roughening of conidia. This placing is based upon his "considerable fruiting mass" hence is little better than a guess. See key to Section Velutina.

34. *C. sormanii* Carbone. Atti Ist. Bot. dell'Universita Pavia, Ser. II, 14, pp. 290-295, 321, Tav. XII. figs. 2, 3, 4.

Translation (C. T.) of Latin diagnosis: "Colonies glaucescent; sterile hyphae hyaline, few septate, 1 $\mu$  diam.; fertile hyphae (conidiophores) simple or scantily branched, hyaline, few septate, slightly attenuate at base, 132 by 1.5 $\mu$ , with apex not or slightly or broadly dilated into a vesicle at the largest 7.2 $\mu$  in diameter and 13.6 $\mu$  in length; conidial fruit green 38 $\mu$  in diameter, 77 to 154 $\mu$  long; sterigmata 3 to 6 in the verticil, hyaline, cylindrical, 7 by 2.5 $\mu$ ; conidia in long chains, greenish smooth, subglobose 2 $\mu$  diam."

Habitat: Upon sausages.

Carbone's figures are obviously schematic and are deemed misleading since the form and relative measurements of conidia and sterigmata are

ignored throughout. Apparently Carbone failed to understand the process of conidium production. Unless some one rediscovers this form and determines its relationship more accurately it may be best to follow Pollacci in placing it here.

35. *P. Trzebinskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat., Ser. B, 1927, pp. 498, 499; Taf. 58; Zaleski no. 375.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, fairly rapidly growing becoming 35 to 40 mm. in diameter in twelve days, liquefying the gelatine slowly but completely, velvety or slightly subfloccose, showing traces of zonation only in the outer areas, within thrown into wrinkles cerebriform in the center becoming radiate, broad, distant and progressively shallower outwardly; in color conidial areas near the margin blue green CdC nos. 378B, 428A, 428B, becoming green 346, 347, 343, 318, with the green fading out in age and the colonies in dark grays tinged with orange yellow such as 194, 198; reverse in pale orange or orange yellow shades such as CdC nos. 153C, 146, 128D, 128C, 121, 122; odor none; conidiophores 100, 200 to 400 or up to 600 $\mu$ , by 2.5 to 3 $\mu$ , with apices commonly inflated 4.5 to 6 or 8 $\mu$ , more or less upright, unbranched, sometimes slightly enlarged upward; sterigmata about 10 to 11 by 2.5 to 3 $\mu$ , in verticils of 6, 10 to 20, or 25; conidia about 2.5 to 3.5 $\mu$ , smooth, globose or subglobose, showing connectives in the chains.

Habitat: Species isolated from earth under pines in "Długa Goslina" near Poznan in Poland. Zaleski classes this form with the "*Aspergilloides* Wehmer-Dierckx Series 4: *D. Stipes acrocarp.* non-divisus Biourge."

Colonies upon Czapek's solution agar spreading slowly, with a thinly developed superficial network of hyphae, bearing part of the conidiophores, with margin plane, thin, fimbriate, 3 to 4 mm. wide with central area variously buckled and wrinkled, with conidial areas in shades of bluish green to green, then gray; reverse becoming dark reddish brown; conidiophores about 2 $\mu$  in diameter; penicilli monoverticillate with conidial chains in a loose column; sterigmata about 9 to 11 by 2 $\mu$ , tapering to rather narrow tubes; conidia elliptical to subglobose or globose, 2.5 to 3.5 $\mu$ , smooth or faintly spinulose.

Culture no. 5010.27 received as type from Baarn in July, 1928, complies fully enough with Zaleski's description to be accepted. Our own culture no. 5016.10 isolated from a rotting *Strobilomyces* is very near to Zaleski's species.

*Species probably belonging here but with descriptions too meagre for identification.*

36. *Citromyces citricus* Mazé and Perrier. Ann. Inst. Pasteur 18:558-559. 1904.

Species obtained from colonies developed upon solutions containing 25 per cent of citric acid, producing in cultures a heavy felt, scarcely floating, or tending to sink when grown upon fluid in containers of large diameter, usually clear slate gray, but becoming deep green when citric acid appears in mineral salt medium as a result of fermentation or if citric acid is introduced as the only source of carbon. Cultures in meat bouillon with sugar added remain white whether citric acid is present or absent, produce long aerial hyphae. No description of sporulation was given. *C. citricus* is closely related to *C. tartaricus* Mazé and Perrier.

37. *C. lacticus* Mazé and Perrier. Ann. Inst. Pasteur 18:558-559. 1904.

Species obtained from colonies developed upon a solution containing 25 per cent of tartaric acid, forming a thin much wrinkled felt upon the surface of fluid media, producing very short aerial hyphae, and becoming ashy or slate blue with the development of the powdery conidia. No description of sporulation was given. The species is closely related to *C. oxalicus* Mazé and Perrier.

The Centralbureau at Baarn gives "*P. lacteum* Mazé et Perrier" in its 1929 list of cultures available. This is probably intended for *C. lacticus*.

38. *C. oxalicus* Mazé and Perrier. Ann. Inst. Pasteur 18:558-559. 1904.

Species obtained from colonies developed upon a saturated aqueous solution of oxalic acid, forming a thin much wrinkled felt upon the surface of fluid media, producing very short aerial hyphae, and becoming ashy or slate blue with the development of the powdery conidia;

Closely related to *C. lacticus* Mazé and Perrier; probably unidentifiable.

39. *C. tartaricus* Mazé and Perrier. Ann. Inst. Pasteur 18:558-559. 1904.

Species obtained from colonies developed upon a solution containing 25 per cent of tartaric acid, producing in cultures a heavy felt, scarcely

floating or tending to skin when grown upon fluid in large containers of large diameter, usually clear slate gray, but becoming deep green when tartaric acid appears in mineral salt medium as a result of fermentation or if tartaric acid is introduced as the only source of carbon; cultures in meat bouillon with sugar added remain white whether tartaric acid is present or absent, and produce long aerial hyphae.

*C. tartricus* is closely related to *C. citricus* Mazé and Perrier.

*Subsection 3. Stricta-funiculosa: colonies producing part or all their conidiophores as branches from trailing or ascending hyphae and interlacing ropes of hyphae. Species varying from floccose in appearance to velutinous with trailing hyphae. (Fig. 22).*



FIG. 22. Diagrammatic representation of monoverticillate aerial ropes of hyphae representing the general type of structure found in the series.

Biourge figures *P. hypo-janthinum* as belonging in this subsection except for special cells figured in the hyphae. His cultures received (see Chapter XXVI) in July, 1928, were labeled correctly as *Aspergillus*.

*Ropiness well marked.*

*Conidia elliptical.*

40. *P. carmino-violaceum* Dierckx. Soc. Scientifique Bruxelles 25: 86, 1901, in Biourge Monogr. La Cellule 33: fasc. 1, pp. 281-282; Col. Pl. X, Cart. 178; Pl. XVI, fig. 93. 1923.

Colonies in wort gelatine restricted in growth, commonly velvety in appearance but with a felt of creeping hyphae and ropes of hyphae, bluish gray green, to brown, with secondary rosy spots or areas from overgrowth of hyphae from the submerged mycelium; coremia none; reverse at first yellowish, then orange, red orange, or violet red; odor none; conidiophores 2 to 3.5 $\mu$  in diameter arising from ropes of hyphae; penicillus 10 to 15 or 30 to 50 $\mu$ , with all walls smooth, figured as simple verticils and branches (? metulae) when present as diverging and

independent rather than parts of a penicillus since main axis and branches are septate and without marks suggestive of a single fruiting structure; metulae 13 to 18 by  $2.5\mu$ , single or few; sterigmata 7 to 10 by 2 to  $3\mu$  in verticils of three to many; conidia ovate 3 by 2, soon 4 by 2.8, finally  $5.5$  by  $4\mu$ .

Biourge's no. 176 (our no. 4733.28) grew on Czapek's solution agar as a wrinkled felt with surface growth consisting of tangled anastomosing networks and ropes of hyphae, colorless to gray becoming purplish brown; reverse becoming purplish brown; drops vinaceous; conidiophores about 100 by  $2\mu$ ; penicillus a single verticil of metulae with occasional sterigmata at the next lower septum; conidia in our cultures elliptical to globose with long axis 2 to  $3\mu$ , in chains parallel or almost in columns at first then tangled, about  $50\mu$  long.

Culture no. 4733.83 received from Biourge (as his no. 375) in September, 1927, labeled *P. lividum* Westling is very close to 4733.28 (Biourge's no. 176). It is certainly not Westling's species and may be regarded as a strain of *P. carmino-violaceum*.

If Biourge's figure and description are correct, his species belongs at this place in the scheme of classification. The measurements he gives for the conidia suggest an *Aspergillus*. Our transfer received from him has conidia which would place it among globose species immediately following.

41. *P. chermesinum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 284-285; Col. Pl. X, Cart. 114; Pl. XVI, fig. 95. 1923.

Colonies on wort gelatine restricted in growth, with margin white, crenulate, 0.5 to 1 mm. broad, bright green, then gray olive, and finally brown; coremia none; reverse orange yellow to purplish red shades to dark reddish brown; odor none; conidiophores 1.5 to  $2.5\mu$  in diameter, arising from creeping hyphae; penicillus figured as simple crowded verticils of sterigmata on unbranched stalks, or duplicated by divergent branches of varying length at varying distances from the verticil on the main axis; metulae (or branches C.T.) very diverse 8 to 25 to 40 to  $65\mu$  long, in pairs or single; sterigmata 8 to 12 by 2 to  $3.5\mu$ , frequently incurved, in groups of 3 to 10; conidia elliptical 2.5 to 4 by 1.5 to  $2.5\mu$ , with ultimate and penultimate ones 5 by  $3\mu$ .

Biourge's type no. 114 (our no. 4733.29) was obtained from Holland cheese. Grown in Czapek's solution agar, no. 4733.29 produced colonies closely felted with more or less fasciculate hyphae, buckled and wrinkled, up to 400 or  $500\mu$  deep, fruiting to the very edge with radiating marginal

hyphae appearing almost velvety at the very edge which is almost white; reverse in gray to orange shades at first then deep purplish red, or deep purple and agar in pale shades of the same colors; microscopic findings as given by Biourge.

42. *P. aureo-flavum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 299-301; Col. Pl. VII, Cart. 56; Pl. XII, fig. 69; 1923.

Colonies on wort gelatine felted, restricted in growth, wrinkled, at first bright green, then gray green, fuscous green and finally reddish brown; coremia none; reverse yellow to orange or orange red; gelatine liquefied, uncolored; odor none; conidiophores 1.8 to 3.2 $\mu$  in diameter, unbranched or very rarely branched; sterigmata 9 to 13 by 2 to 3.2 $\mu$ , occasionally to 4 $\mu$ , in groups of 2 to 6; conidia elliptical 2.5 to 4 by 1.5 to 2.5 $\mu$ , with terminal conidium of the chain 4.4 $\mu$  almost globose.

Biourge's type no. 56 was not received. In September, 1927, we received from Biourge a contaminated culture labeled *P. aurantio-flavum* (4733.6.1) which contained a *Penicillium* closely enough complying with the description of *P. aureo-flavum* to suggest that the label was a mistake in copying.

*Stricta-funiculosa.* 2. *Conidia globose.*

45. *P. Niklewskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 504, 505; Taf. 60; Zaleski no. 1366b.

Colonies upon neutral Raulin with 10 per cent gelatin in petri dishes, slowly growing, becoming 25 to 28 mm. in diameter in twelve days, liquefying the gelatine only slightly and tardily, thin, with superficial growth of trailing and anastomosing hyphae and ropes of hyphae, zonation more or less evident, with central area more or less radiately wrinkled; marginal zone 1.5 to 2 mm. wide white; in color conidial areas at the margin blue green shades such as 378B and C., then 372, and green 347, with the green fading in age giving dark orange gray shades such as 139, 143; reverse in light yellow shades such as 203B, 171, 166; odor none; conidiophores 10 to 150 or up to 300 $\mu$  by 2 to 2.5 $\mu$ , unbranched straight, or flexuous erect or ascending, commonly enlarging upwards to a more or less definite vesicle, commonly arising from ropes of hyphae; sterigmata about 9 to 10 by 2.3 to 2.8 $\mu$ , in verticils of 4, 8 to 20, or up to 25, figured as more or less diverging, described as with marginal sterigmata incurved; conidia 2 to 2.5 $\mu$ , smooth, globose, showing connectives in the growing chains.

Habitat: Species isolated from earth under conifers in Poland.

Zaleski notes that Biourge agrees that this is a new species and belongs in "Aspergilloides Wehmer-Dierckx Series 4: D. Stipes acrocarp; non-divisus Biourge."

Type strain growing better at 30° than at 20°C.; colonies upon Czapek's solution agar fairly broadly spreading, plane or with shallow radiate wrinkles, floccose with bristly prostrate and ascending ropes or coremia of hyphae, making a mass 1 to 2 mm. deep, (upon wort the same structures enlarged, almost erect), becoming slowly pale green or gray green; reverse not colored at first, later gray and finally showing dark zones; odor, none; conidiophores mostly short branches (up to 20 by  $2\mu$ ) from the funiculose or fasciculate bundles of hyphae; penicilli consisting of single verticils of sterigmata crowded at base, diverging at apex and bearing diverging conidial chains; sterigmata about  $7\mu$  long; conidia 2 to  $2.5\mu$  in diameter, smooth, globose with connectives prominent.

Culture no. 5015.17 received from Baarn in July, 1928, appears to be type.

46. *P. Adametzi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 507, 508, 509; Taf. 47, 61; Zaleski no. 1041.

Colonies in neutral Raulin with 10 per cent gelatin in petri dishes, slowly growing becoming 25 to 27 mm. in diameter in twelve days, liquefying gelatine slowly but completely, azonate, with thallus in radiate wrinkles, and surface growth consisting of anastomosing hyphae and ropes of hyphae, which become coremia in acid media; conidial areas in blue green shades such as CdC nos. 378B, 371, 372, 373, 367, to green 346, fading to shades of gray; margin white 2 to 3 mm. wide in the growing colony; crystals abundantly produced in media initially acid in reaction; drops small yellow, abundant in the furrows toward the margin; reverse pale orange yellow shades such as 153c, 171, 196; odor none; conidiophores 30, 40 to 80 or up to  $100\mu$  by 2 to  $2.3\mu$ , more or less inflated at the apex, straight or slightly flexuous, rarely branched, arising from trailing hyphae or ropes of hyphae; sterigmata about 7 to 8 by 2 to  $2.3\mu$  in compact verticils of 4, 8 to 12, or up to 16 with short tubes; conidia about 2 to  $2.3\mu$ , smooth, globose, fairly uniform in size and shape, showing connectives.

Habitat: Species isolated from earth under conifers near Poznan in Poland.

Zaleski placed this species in "Aspergilloides Wehmer-Dierckx Series C: Stipes acrocarp. non-divisus Biourge."

Colonies in Czapek's solution agar growing well up to 30°C., forming close woven felts with almost velvety appearance but consisting of hyphae and fasciculate or funiculose hyphae interlacing, radiately wrinkled, with central area buckled; in color in the central area, white, the intermediate area pale green, and an outer broad (about 5 mm.) band white, with marked zonation appearing in the outer areas in old cultures (azonate when young); drops abundant when grown at 20 to 22°C., less evident at 30°C. reverse pale yellow to brownish, with prominent radiate wrinkles; agar more or less colored in slants in age; penicilli monoverticillate or with an occasional branch, more rarely 2 to 3

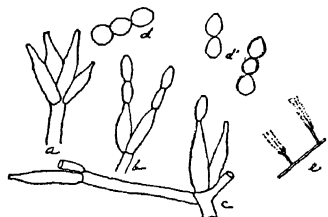


FIG. 23

FIG. 23. *P. vinaceum* Gilman and Abbott: *a*, *b*, *c*, types of verticils of sterigmata with one sterigma prolonged into a branch bearing a secondary verticil in *c*; *d*, ripe conidia nearly globose; *d'*, developing conidia; *e*, habit sketch from Gilman and Abbott.



FIG. 24

FIG. 24. *P. decumbens* Thom: *ab*, surface of substratum; *sm*, submerged hyphae; *t*, trailing hypha bearing conidiophores.

branches with secondary penicilli, or occasionally superposed verticils of sterigmata; sterigmata 7 to 9 $\mu$  by 2 to 3 $\mu$ , closely packed at base, diverging at apex, few to many in the verticil, bearing divergent chains of conidia; conidia colorless, globose, smooth, 2 to 2.5 $\mu$  in diameter.

Culture (5010.1) received from Baarn in July, 1928, appears to be type.

47. *P. vinaceum*. Gilman and Abbott. Iowa State College Jour. Sci. 1; no. 3, p. 299, fig. 34. 1927. Type culture Thom and Church no. 4894.15, fig. 23.

Colony upon Czapek's solution agar after thirteen days showing a heavy felt 4 cm. in diameter and varying up to 3 or even 4 mm. deep; radiately wrinkled and buckled with margin crenulate and producing heavy floccose masses several millimeters deep in age; consisting of



delicate hyphae  $2\mu$  or less in diameter tearing fairly easily and lifting the agar with the mass, separate and fasciculate woven into felts; margin heavy white felt with aerial hyphae to the very edge; central area vinaceous uneven with vinaceous drops and vinaceous fluid readily squeezed out; green color absent or reduced to a tan in the more crowded conidial areas, dark vinaceous (R. XXVII.1<sup>11</sup>) with color broadly diffused in age; reverse vinaceous; adequately colored vinaceous throughout; odor, none; penicilli borne on short stalks from floccose hyphae, mostly monoverticillate or occasionally a mixed verticil of sterigmata and proliferated members with secondary heads, or even occasionally producing diverging branches as in the soil *Penicillia*; conidia when ripe, 3, mostly 2 to  $2.5\mu$  in long axis in earlier stages; apiculate at both ends while developing, becoming more or less completely globose when ripe.

Habitat: Soil.

48. *C. griseus* Sopp. Monogr., pp. 119-120, Taf. XV, fig. 104; Taf. XXII, fig. 5. 1912.

Colonies in meat-peptone-sugar-gelatine producing close felted but thin, tough, wrinkled, paperlike mycelium, gray green to bluish green or mouse-gray, in age brown; reverse chamois red (deep orange); odor weak or doubtful; conidiophores erect, short, slender, septate, often with one or more diverging branches, without vesicular apex, figured as if partly arising from trailing hyphae (?); sterigmata small, short and figured as few in the verticil; conidia (from Sopp's tabulation not given in description) 2.5 to  $3\mu$ , globose, smooth; perithecia not found.

Species found in earth in East Norway; colonies grew from  $1^{\circ}\text{C}$ . to  $40^{\circ}\text{C}$ . with optimum between 30 and  $35^{\circ}\text{C}$ . At very low temperatures structures suggestive of perithecia were produced. Good growth was produced in milk, urine, meat-peptone broth, wort, potato, and bread. Little odor, small enzymic and fermentative activities, and no coloring substances were detected. Conidia survived more than three years in the laboratory.

No one has been able to identify this species since it was described.

49. *P. platense* Spegazzini. Rev. Agr. y. Veter. La Plata (1896), p. 246.

Colonies upon leaf sheaths and internodes of cane, subhemispherical 350 to  $500\mu$  in diameter, erumpent to superficial, more or less densely grouped, glaucous green, with vegetative hyphae partly creeping, partly penetrating into the substratum; conidiophores 100 to  $150\mu$

long by  $3.5\mu$  in diameter, arising as branches from fascicles or ropes of aerial hyphae, not or scarcely enlarged at the apex, bearing 7 to 12 branches (sterigmata), 20 by  $3.35\mu$ , attenuate at the apex and bearing long chains of conidia; conidia globose, 2 to  $2.5\mu$  diameter, pale green, smooth.

Reported only upon herbarium materials consisting of leaf sheaths and internodes of cane from La Florida and La Plata in 1894.

*\*Colonies velvety in appearance, but with gradation from trailing hyphae to trailing ropes of hyphae.*

50. *P. decumbens* Thom. In U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118: p. 71, fig. 28. 1910. See fig. 24. See Biourge Monogr. La Cellule 33: fasc. 1, pp. 287, 288; Col. Pl. IX, Cart. 110; Pl. XV, fig. 89. 1923.

Cultivated in gelatin or potato agar, white to gray, gray-green ultimately yellowish brown, green in cultures with cane sugar; surface growth consisting of trailing or stolon-like hyphae sparsely developed and so close to the substratum as to appear only as fertile hyphae, bearing the conidiophores as short branches 20 to  $100\mu$  in length, in old colonies with dense tufts of sterile secondary mycelium scattered upon the surface; conidial fructifications consisting of single verticils of crowded sterigmata cells, 7 to 9 by 2 to  $3\mu$ , bearing conidial chains first in loose columns up to  $100\mu$  in length but upon potato agar soon becoming enveloped and broken up in the drops of fluid (*Gliocladium*-like); conidia globose 2.5 to  $3\mu$ , vacuolate, smooth, pale green then brownish in mass; colonies do not liquefy gelatin; give a weakly alkaline reaction to litmus; produce a definite odor in cultures containing cane sugar.

Contributed by Prof. P. H. Rolfs from Miami, Fla. 1905.

In Czapek agar in petri dishes, colonies are velvety slowly but fairly broadly spreading, plane, 100 to  $200\mu$  deep, and show faintly a narrow zonation, with outer margin white, a band of light drab within and central areas in drab shades (Ridgway XLVI). Microscopic structures as already described except that the conidia persist in chains.

The original culture of *P. decumbens* in our collection has been lost; Biourge, however, returned to us his culture no. 110 (our 4733.49) received by him from Kral which is apparently identical with the original sent by us to Kral then in Prague. This organism was isolated in Miami, Florida, by Prof. P. H. Rolfs in 1905. In addition, the following

cultures reproduce the morphology and reactions of the original, from Cape Town, South Africa, isolated by Putterill 4648.36.3 and .161.2; from wood, isolated in the Forest Products Laboratory at Madison, Wis., 4312.71218.2; from D. H. Linder, 4754.C79.

51. *P. citreo-sulfuratum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 285–287, Col. Pl. IX, Cart. 21; pl. XV, fig. 86. 1923.

Cited as in Dierckx MSS. (unpublished) as *P. citreo-nigrum* var *sulfurea*.

Colonies on wort gelatine restricted in growth, more or less wrinkled, more or less evanescently bluish green, then grayish sulfur yellow, at length reddish brown; coremia none; reverse more or less citrine yellow; odor, none; conidiophores up to 100 or more by  $2\mu$  borne upon trailing, creeping or ascending (rampant) hyphae, with all walls smooth; penicillus figured as a single verticil of sterigmata, or a main axis with one divergent branch (metula?) at the topmost node, with occasionally another lower down; branches 8 to  $30\mu$  in length; sterigmata 5.5 to 7.5 occasionally to 11 by 2 to  $2.5\mu$ , in groups of 2 to 10, figured as so grouped that they diverge at the apex which tapers toward a fairly fine point; conidia subglobose 2.8 by 2.2, or 2 to  $3\mu$ .

Biourge type no. 21 was not received by us. The name was apparently based upon sulfur yellow in reverse and agar and present also in margin or in aerial vegetative hyphae.

52. *P. fellutanum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 262–264; Col. Pl. XIII, Cart. 177; Pl. XXIII, fig. 133. 1923. See fig. 25.

Colonies on wort gelatine restricted in growth, at first transiently bluish green, then gray olive; coremia none; reverse at first pale yellow, then sordid rosy; odor weak; conidiophores rising from creeping hyphae 10 to 35 by 2 to  $3.5\mu$ ; penicillus 20 to  $35\mu$  long, with all walls smooth, figures variously from monoverticillate (Citromyces-like) to a dense group of metulae, always on short stalks from creeping hyphae; metulae 9 to 13 or even to 20 by 2 to  $3\mu$ , in groups of 2 to 5; sterigmata 6–11 by 1.5 to  $4\mu$ , in verticils of 2 to 8; conidia oblong 2 to 3 by 1.8 to  $2.5\mu$ .

Biourge's type culture no. 177 (our no. 4733.58) was obtained from a skim milk cheese at Feluy (Fellut in Celtic).

Two strains coming to us under the name have been studied—Biourge's no. 177 (our no. 4733.58) as received was a mixture from which we separated a green form probably representing Biourge's species.

The other, no. 4876.24 from Dr. Westerdijk, varied somewhat in details but is believed to be the same species; colonies upon Czapek's solution agar narrowly growing, with margin velvety, and not over 100 to 200 $\mu$  deep showing radiating lines of conidial areas running out unevenly, with central areas thin almost papery but convoluted, or wrinkled with ridges sometimes 1 to 2 mm. high, in color various shades of bluish green, or yellowish green with faintly bluish effect, becoming gray in age, reverse in yellowish or yellowish green, sometimes sordid rosy, again shading toward dark almost black areas, with the wrinkling of the colony distinctly evident; conidiophores usually less than 100 $\mu$  long, either arising from a closely woven felt, or from trailing or prostrate creeping hyphae; penicillus mostly monoverticillate with conidial chains adherent into a column up to 100 $\mu$  long, or, sometimes apparently coalescing in a mass almost Gliocladium-like, at times with secondary

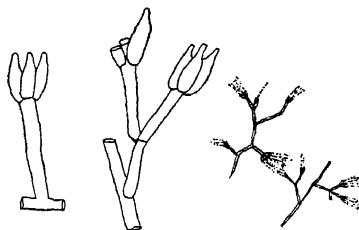


FIG. 25. *P. fellutanum* Biourge: Showing the branching type of trailing hyphae bearing conidiophores.

and usually divergent columns arising from single or asymmetrically verticillate branches or metulae; serigmata 6 to 8 $\mu$  long in 4876.24, 8 to 12 $\mu$  in 4733.58, closely packed in the verticil, usually few; conidia varying 2.5 to 3 by 2 to 2.3 $\mu$ , or subglobose about 2.5 $\mu$ , with occasional larger ones up to 4 $\mu$  which may be in the early stages of germination, almost colorless under the microscope.

53. *P. citreo-viride* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 297-299; Col. Pl. IX, Cart. 58; Pl. XV, fig. 88. 1923.

Colonies in wort gelatine, undulate, wrinkled, forming a thin fibrous felt, pale bluish green, quickly passing to sordid dark violaceous shades at length subfuscous; coremia none; reverse at first white then pale yellow to greenish yellow, or orange; odor none; conidiophores 25 to 80 or longer by 1.5 to 2 $\mu$ , arising from creeping hyphae; penicillus very

short as a simple verticil of sterigmata, or a main axis with sterigmata in a terminal verticil or in sessile or stalked groups from lower nodes; sterigmata 6 to 9 by 1.5 to  $3\mu$ , occasionally much larger, in apical verticils of 2 to 10, at lower nodes singly or clustered or on true metulae occasionally; conidia globose 2 to  $3\mu$ .

Biourge's type no. 58 (our no. 4733.38) on Czapek's solution agar produced colonies rather small, restricted in growth, wrinkled, buckled, umbonate in center, velvety appearing but with a thin mycelial felt in all about 100 to  $200\mu$  deep, thinning to a fibrous white margin, with the conidial areas pale bluish green then pale green becoming brown shades in the older central area; reverse pale yellow to greenish or greenish-yellow; agar pale yellow; hyphae very delicate; conidiophores about  $2\mu$  in diameter, producing a simple terminal verticil of sterigmata, or with central axis prolonged to form a second verticil, or with 1 or 2 branches at the first and second nodes, or with sterigmata and metulae in the same verticil; sterigmata 7 to 12 by 1.5 to  $2\mu$ ; with points slender; conidia about  $2.5\mu$  in diameter, with chains aggregated into a tangled but more or less columnar mass.

Biourge assigned the species to his *Thyrsiferes*—series *Citrinum*. Our cultures verify Biourge's description, but seem to justify placing it here.

Biourge's no. 98 (our 4733.84) was received in September, 1927, labeled *P. luteo-viride* but when cultivated checked up with his *P. citreo-viride*, not with his description of *P. luteo-viride*. Bainier and Sartory figured *Citromyces minutus* with structures which might easily be placed here.

An occasional form met in miscellaneous culture work seems to justify retaining this species.

54. *P. phaeo-janthinellum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 289–290; Col. Pl. VIII, Cart 35; Pl. XIII, fig. 77. 1923.  
See also, Van Beyma Thoe Kingma, Verhandel. Kon. Akad. Wetensch. Amsterdam Tweedie Sec. 26, no. 2, p. 21. 1928.

Colonies in wort gelatine, appearing velvety ("raso") but showing a felt of aerial hyphae bearing the conidiophores, wrinkled, obscurely blue green, then obscurely rosy, at length fuscous; conidia none; reverse from greenish to orange shades to purplish violet; odor weak; conidiophores 20 to 30 by 2 to  $3\mu$  arising from creeping hyphae or ropes of hyphae; penicillus 10 to  $25\mu$  long, with all walls smooth, figured as simple monoverticillate forms or pairs of unequal diverging branches

or metulae; metulae 10 to 20 by 1.5 to 2.8 $\mu$ , in pairs or single; sterigmata 6 to 8 by 1.8 to 2.4 $\mu$  in fours or fives, figured as producing parallel diverging chains of conidia; conidia subglobose 1.5 to 3 $\mu$ .

Biourge's type no. 35 (our no. 4733.96) upon Czapek's solution agar produced colonies, spreading, with surface apparently velvety but from a thin close lying felt with prostrate anastomosing ropes of hyphae at the surface of the agar, in all 100 to 200 $\mu$  deep, with wrinkles, buckling, and some overgrowth in center; gray to very pale green, reverse colorless or slightly greenish to dark greenish color unevenly distributed; conidiophores up to 50 $\mu$  long with terminal verticils of few sterigmata and chains of conidia forming short columns.

55. *P. cinerascens* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 308-309; Col. Pl. IX, Cart. 50; Pl. XIV, fig. 81. 1923.

Colonies on wort gelatine, restricted in growth, with trailing or ascending hyphae, at first slightly bluish green then gray green to gray at length reddish brown (hyacinth); coremia none; reverse pale yellow; gelatine liquefied but not discolored; odor, none; conidiophores smooth, about 35 $\mu$  long and 1.5 to 2.8 $\mu$  in diameter, only slightly swollen at the apex, arising from ascending (aerial) hyphae; sterigmata commonly gibbous, attenuate at both ends, 6 to 11.5 by 1.5 to 2.8 $\mu$ , in verticils of 4 to 10, smooth, rather divergent; conidia oblong to globose, 2-3.8 by 2.2 to 3 $\mu$ , rough echinulate when ripe and showing the "Phenomenon of Corda," the end cell much larger than the others in the chain; Biourge type no. 50 was not received.

Type 4733.34 received in September, 1927, grown slanted tubes of Czapek's solution agar showing salmon to pinkish tint in reverse; in petri dish, almost or completely colorless below; colonies 1 to 2 cm. in diameter; a close floccose felt of delicate hyphae; white to pale gray, slowly developing pale green marginal zones passing through gray to white in center or at times showing a faint greenish tinge; hyphae delicate; conidiophores partly branches of aerial hyphae 20 to ? $\mu$  long; partly rising separately up to 300 to 400 by 2 $\mu$ ; penicillus consisting of 1 verticil, with chains parallel, fairly closely or partly adherent in columns up to 100 $\mu$  or 150 $\mu$ ; sterigmata 6 to 7 (8 $\mu$ ) long by 1.5 to 2 $\mu$ ; conidia elliptical to globose up to 2 $\mu$  or a little longer in long axis.

*P. cinerascens* in Gilman and Abbott (No. 4894.12) as received does not belong here but among the "Divaricata" near their *P. guttulosum*.

Another culture, no. 5037.109, received from the British Cotton Industry Research Association seems close enough to this description

to be assigned to *P. cinerascens*: Our notes follow: Colonies upon Czapek's solution agar forming close textured felts of fine hyphae, with rosy or funiculose masses of hyphae 300 to 400 $\mu$  deep in the central area becoming much deeper in the raised marginal zone as the colonies became older, in color gray or greenish scarcely green; reverse colorless to pale yellow, pale orange shades; drops crystal; conidiophores usually not over 40 $\mu$  long, borne as short branches from masses or ropes of hyphae, broadening toward a vesiculose apex sometimes gradually from base to apex, sometimes with apex 1 to several branched to produce a divergent verticil, which is apparently due to the development of the sterigmata into verticils of metulae; sterigmata 7, 8 or sometimes 10 $\mu$  by 2 to 2.5 $\mu$ ; conidia 3 to 3.5 by 2 to 2.5 $\mu$ , mostly very small 2 to 2.5 by 1.5 $\mu$  in the earlier or growing stages of the colony.

56. *P. Paczoskii* Zaleski. In Bul. Acad Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 505, 506, 507; Taf. 47, 61; Zaleski no. 1609b.

Colonies upon neutral Raulin with 10 per cent gelatine in petri dishes, fairly rapidly growing becoming 38 to 42 mm. in diameter in twelve days, liquefying the gelatine slowly but completely, thin, plane, velvety-figured as with trailing hyphae and ropes of hyphae, zonate, more or less radiate wrinkled within and more or less overgrown with secondary mycelium in center; with a marginal band 6 to 8 mm. wide white or with faint bluish color, central areas bluish green CdC nos. 372, 422, 423, 375, 396, losing the blue green in age and becoming yellowish gray shades such as 164, 160; drops numerous in the intermediate zones, uncolored; reverse in pale orange shades such as CdC nos. 146, 121, 97, 128A; odor none; conidiophores 50, 100 to 200 or up to 300 by 2.2 by 2.5 or 3 $\mu$ , commonly inflated at apex to 4 to 5 $\mu$ , arising largely from coarse ropes of hyphae, mostly unbranched, straight or flexuous; sterigmata about 9 to 10 by 2 to 2.5 $\mu$ , in crowded verticils of 8, 10 to 15 or 20 with short tubes; conidia 2.5 to 3 $\mu$ , or up to 3.5 $\mu$ , smooth, subglobose to ovate, showing connectives in the chains.

Habitat: Species isolated from earth under conifers in the forests of Poland.

Zaleski found difficulty in getting characteristic conidium producing colonies only once apparently obtaining the data recorded. He placed the species in "Aspergilloides Wehmer-Dierckx Series 4: D. Stipes acrocarp. non-divisus Biourge." Our notes follow: Type strain growing more slowly at 30° than at 20°C.; colonies upon Czapek's solution agar at 20°C., plane, broadly spreading, becoming 30 to 35 mm. in

diameter in seven days, sometimes radiately wrinkled, slowly zonate with scanty superficial trailing hyphae and occasional ropes of hyphae small, prostrate or rising only slightly above the substratum, visible mostly in the marginal area; conidial areas developing as raised velvety cushions transiently bluish green then dusky olive green with an uncolored margin 5 to 6 mm. wide consisting partly of aerial, partly of submerged hyphae; reverse colorless to pale yellow; conidiophores up to  $100\mu$  or  $150\mu$  by about  $2\mu$  mostly arising from submerged hyphae; penicilli consisting of single verticils of sterigmata bearing chains of conidia parallel or forming a loose column up to  $200\mu$  long, the chains more or less separating and becoming tangled in age; sterigmata few in the verticil, about 10 by  $2\mu$ ; conidia up to  $3\mu$  less often  $3.5\mu$ , smooth as seen with low magnification, very delicately punctate as seen under oil immersion.

Culture no. 5010.18 received from Baarn in July, 1928, appears to be type. A closely similar strain (no. 4718) was isolated by us from moldy rubber.

57. *P. terlikowskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 501, 502; Taf. 59; Zaleski no. 392.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, fairly rapidly growing becoming 32 to 36 mm. in diameter in 12 days, liquefying gelatine, velvety, usually but not always zonate, with the central area in cerebriform wrinkles changing to radiate in the intermediate area; marginal zone rosy; abundant crystals produced in acid media; in color conidial areas near the margin in blue green shades such as CdC nos. 378A, 378B, becoming 367 or green 342, 318, 319, within, and in age dark orange shades such as 134, 143; reverse orange yellow and orange shades such as 196, 157, 166, 171, 109, 110; odor none; conidiophores 100 to 150 or up to  $250\mu$  by  $2.5$  to  $3\mu$ , with or without inflation of the apices to 4 to 5 or  $6\mu$ ; sterigmata about 9 to 11 by  $2.2$  to  $2.5$ , in compact verticils of 3, 8 to 12, or up to 15, with short tubes; conidia  $2.2$  to  $2.5$  or  $3\mu$ , smooth, more or less globose, showing connectives distinctly.

Habitat: Species isolated from earth under conifers in a forest near Poznan in Poland.

Zaleski notes that his cultures of this species showed colorless drops but his later transfers failed to produce such drops hence all reference to them was dropped from the description. He lists it among the "Aspergilloides Wehmer-Dierckx Series 4: D. Stipes acrocarp: non-



divisus Biourge." Our notes follow: Colonies in Czapek's solution agar spreading rather broadly, mostly submerged and colorless or pale flesh color becoming greenish slowly beginning in the central area, rather broadly zonate, with fairly wide submerged margin, within broad band with sparse white aerial hyphae and a central area green, characterized by a mixture of conidiophores rising from submerged hyphae and as branches from a loose fibrous network of hyphae and ropes of hyphae, radiately wrinkled; reverse colorless at margin, fairly bright yellow to deep ochraceous orange; conidiophores about 2.5 to 3 $\mu$  in diameter, and 100 $\mu$  long when arising from submerged hyphae, up to 50 $\mu$  long as branches of aerial hyphae, with walls pitted or roughened; penicilli consisting of a single closely packed verticil of sterigmata producing a column of conidial chains 100 to 150 $\mu$  long; sterigmata 8 to 9 $\mu$  long; conidia subglobose 2.5 to 3 $\mu$  faintly pitted or roughened.

Culture no. 5010.25 received as type from Baarn in July, 1928, complies with Zaleski's description well enough to be accepted.

58. *P. roseo-cinnabarinum* Biourge. Monogr. La Cellule 33: fasc. 11. pp. 319-321; Col. Pl. X, Cart. 3r; Pl. XVII, fig. 97; 1923.

Colonies in wort gelatine, restricted in growth; for the most part rosy in color, rarely with blue green areas; with margin broad white or yellow or becoming red; coremia none; reverse in central areas cinnabar, to dark blood red, with margin yellow shading into red; odor, none; conidiophores smooth, 60 to 100 by 2.5 to 3 $\mu$ , with apex clavate or vesicle-like and 7 to 9 $\mu$  in diameter, arising from creeping hyphae, figured as almost Aspergillus-like but failing to show the nature of the basal cell and the cell-relations of the vegetative hyphae; sterigmata mostly 7 to 9 and very numerous, less commonly up to 16 by 2 to 3 $\mu$ ; conidia elliptical 3.5 to 5 by 2.4 to 4 $\mu$ , delicately roughened.

Biourge's type no. 3r was lost by him many years ago. It appeared as a contaminant of a culture of *P. solitum* and was characterized by a rosy surface area producing blue green spores only on bread and upon alkaline bouillon at 20°C. In general appearance this description suggests *P. islandicum* Sopp but the figures and description define a strictly monoverticillate form hence it must be placed arbitrarily until some one rediscovers it and ascertains its real relationship. One is tempted to raise the question suggested by Biourge's notes whether he did not have an Aspergillus in culture rather than a Penicillium when he described this species.

*Subsection 4. Stricta-velutina: colonies velvety.*<sup>2</sup>*\*Conidia elliptical rough or spinulose.*

65. *P. lividum* Westling. Arkiv för Botanik 11, pp. 58, 134–137; fig. 79. 1911. See Dale, E. Ann. Mycol. 12: 52. 1914.

Colonies in prune gelatine slowly growing, floccose, spreading broadly, white then blue-green or gray blue (C.d.C. 422–423, 393, 398), and in very old colonies black brown, white margin broad; reverse uncolored or subflavus; gelatine not liquefied or only slightly softened; odor scarcely recognizable; conidiophores arising from submerged hyphae, smooth, unbranched, or rarely once branched toward the base, from 90 to 450 by 2.2 to 3.5 $\mu$ , bearing a single crowded verticil of sterigmata 9 to 12 by 2 to 2.4 $\mu$ ; conidia elliptical to ovate, smooth or roughish (in our cultures regularly rough.—C.T.) 2.7 to 3 by 2.2 to 2.6 $\mu$ , swelling in germination to 4.5 to 6 $\mu$ .

The type strain was found on a root-stock of *Polystichum filixmas*. Westling lost this culture. Westling reported that his species formed oxalic acid and small amounts of some other acid. It grew well upon the common media.

Our culture no. 2697 was received from Miss Dale (II, p. 52) from Scotland (D4), and identified by us, then sent to Westling who verified the identification. Several cultures belonging to this species have been obtained since in America. Redescription from our own cultures follows:

No. 2697. Colonies on Czapek's solution agar with 3 per cent cane sugar, dark blue green to deep blue (C.d.C. no. 408, 413, to 418 to 389 or according to Westling nos. 422, 423, 393–398), deeply velvety, with narrow margin, spreading slowly; reverse of colony yellow (flavus) to tan (Code nos. 171, 152, 87) in older parts; agar pale yellow, numerous large globose vesicles are formed by swollen cells of the submerged hyphae; conidiophores mostly simple arising from substratum separately, up to 400, even to 600 $\mu$  or longer, 2.5 to 4 $\mu$  in diameter, swelling at the apex to a vesicle which may be 8 $\mu$  in diameter, penicillus usually a single verticil of sterigmata with conidial chains forming loose columns, sometimes a branch somewhat below the tip bears a second mass of conidia; sterigmata 8 to 12 by 2 to 3 $\mu$ , crowded in verticils; conidia at

<sup>2</sup> Certain species notably Zaleski's *P. Trzebinskii* with velvety appearances have been placed in the subsection *floccosa* because of thin basal felts of aerial hyphae and of other affinities with *P. spinulosum*.

first smooth, elliptical, later rough, 2.6 to 3 by 3 to  $4\mu$  in long chains; odor faint or none.

Gelatin in water produced thin colonies of characteristic color, without color below, but with little or no liquefaction showing in 14 days (no liquefaction in a month in one series). Reverse not colored in bean agar.

Among the strains seen we have records as follows: 4055.4 from New Jersey soil; 4732 from Amherst, Mass.; 4285.1 from C. L. Shear; 4400 from Swedish bread (imported).

Biourge (monogr. La Cellule 33: fasc. 1, p. 297; Col. Pl. XI, Cart. 375; Pl. XVIII, fig. 106, 1923, discussed his culture no. 375 in terms which make us doubt his identification. A culture no. 4733.83 received from him in September, 1927, as no. 375, is not *P. lividum* in the sense of Westling, but is closely allied to his *P. carmino-violaceum* Dierckx. It is probable that Biourge did not have *P. lividum* in culture when he wrote his discussion.

66. *P. Dierckxii* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 313-315; Col. Pl. X, Cart. 12; Pl. XVI, fig. 91. 1923.

Colonies in wort gelatine velvety, much wrinkled, at first blue green (C.d.C. 403<sup>D</sup>, 428<sup>C</sup>, 423) then gray green, or pale glaucous, finally gray with more or less reddish brown; coremia none; reverse more or less sordid orange yellow, finally reddish brown, to dark blood red; odor none; conidiophores 30 to 60 by 1.8 to  $2.8\mu$ , with all walls smooth, without enlargement of apex, arising from decumbent hyphae; sterigmata 9 to 13 by 1.8 to  $2.8\mu$  in verticils of 4 to 10; conidia elliptical 2.5 to  $3.8\mu$  by 2 to  $2.4\mu$ , in germination 5 by  $3.8\mu$ .

Biourge type no. 12 (our no. 4733.50) grown in Czapek's solution agar produced colonies restricted in growth, small, narrow bordered, velvety not over  $100\mu$  deep, close textured with a very smooth surface prominently buckled and umbonate in center; Hathi gray to Dawn gray (Ridgway LII. 35'''''); reverse yellow-citrine through shades of vinaceous to reddish brown under the buckled central area; conidiophores arising from looping or trailing hyphae, about 50 by  $2\mu$ ; sterigmata 7 to 10 by  $2\mu$  taper pointed, few to many in the verticil; conidia oval 3 to 4.5 by 2 to  $3\mu$ , smooth, thin walled, very pale colored, in loosely parallel to semidiverging chains forming semicolumnar masses up to  $200\mu$  long, and falling away in loose powdery masses when tapped.

67. *P. turbatum* Westling. Arkiv för Botanik 11, pp. 54, 128–130; fig. 36, 74a and b. 1911.

Colonies in prune gelatine closely velvety in appearance but with a basal network of aerial branches forming thin mycelium, with conidial areas at first green (prasinus, C.d.C. 347) then gray-green (347–372), becoming clear brown only after a month or more, with white margin very narrow; reverse uncolored; gelatine slowly liquefied, beginning in 5 to 6 days, with a neutral or weakly alkaline reaction; odor wanting; conidiophores arising from creeping hyphae usually very short but not over  $120\mu$  long by 3 to  $4.5\mu$  in diameter; penicillus 20 to  $85\mu$  long, either a single verticil of sterigmata or a verticil of metulae each bearing a monoverticillate conidial mass; metulae when present 2 to 3 in the verticil and 12 to 20 by 2.8 to  $4\mu$ ; sterigmata 8 to 10.5 by 2 to  $2.6\mu$ , figured as diverging at the tips, numerous in the verticil; conidia elliptical, smooth 3 to 3.5 by 2.2 to  $2.8\mu$  in germination 4.5 to 6 by 6 to  $7.5\mu$ ; perithecia (= sclerotia?) colorless to yellowish, ovate to globose, 55 to  $105\mu$  in diameter appearing after seven to eight days, becoming numerous and giving a granular look to the colony.

Species found on branches of *Taxus baccata* as a firm white mycelium developing after the twigs had been kept moist some time. Cultures grew well at 30 to  $31^{\circ}\text{C}.$ , also grew well in malt-extract-gelatine, plum agar, sugar solution, bread, potato. In milk few conidia are formed and the fluid becomes clear yellow.

Biourge (Monogr. La Cellule 33: fasc. 1, pp. 277–278; Col. Pl. XI, Cart 378; Pl. XX, fig. 115; 1923) gave his description. Colonies in wort gelatine velvety, thin, zonate, wrinkled, at first blue green, then gray green, at length reddish brown, coremia none; reverse greenish with zones or spots of red or brown; odor none; conidiophore up to 100 by 3 to  $4\mu$ ; penicillus 15 to  $50\mu$  long, with walls smooth; metulae 15 to 30 by 2 to  $3.5\mu$ , single or none; sterigmata 9 to 14 by 2 to  $3\mu$  in verticils of 4 to 7; conidia 3.5 to 4.5 or even 5 by 2 to  $3\mu$ .

Biourge's no. 378 (our no. 4733.122) grown upon Czapek's solution agar produced colonies rather widely spreading, velvety 100 to  $200\mu$  deep, plane in petri dish cultures, or buckled and more or less radiately wrinkled in slanted tubes, in shades of pale yellow-green, tea green to olive gray; in reverse various shades such as sordid orange yellow, vinaceous fawn, light drab; penicilli monoverticillate with chains of conidia partly parallel almost in columns, measurements of sterigmata and conidia as in Biourge's description.

68. *P. aurantio-violaceum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 282-284; Col. Pl. X, Cart. 33; Pl. XVI, fig. 94. 1923.

Colonies on wort gelatine, somewhat restricted in growth, forming undulate serpentine masses, velvety, dark gray green (C.d.C. 339, 343), with margin bluish gray green, in age orange brown with white warts or tubercles; coremia none; reverse obscurely zonate in yellow, pale orange to carmine shades, in age violet or reddish brown colors; odor none; conidiophore 2 to  $2.4\mu$  in diameter with all walls smooth; figured as arising from creeping hyphae; penicillus  $15\mu$  or up to  $45\mu$  if branched; mostly a simple Citromyces, but occasionally with a single branch or metula; metulae 25 to 30 by  $2.4\mu$  in pairs or single, frequently clavate (this means usually a monoverticillate mass but with an occasional branch from the first node—C. T.); sterigmata 8.5 to 13 by 2 to  $2.5\mu$ , frequently incurved, in verticils of 2 to 10; conidia elliptical 3 to  $3.2$  by 2 to  $2.4\mu$ , except the one at the end of the chain  $5.6$  by  $2.8\mu$ , others do not exceed 4 by  $3.2\mu$  even in germination.

Biourge's no. 3 (not received) is recorded as showing the "phenomenon of Corda." (See Chapter VI.) The note, "sterigmata frequently incurved," merely means a closely packed verticil of many sterigmata.

Colonies with this morphology are occasionally found; one of these may be described as follows: Colonies on Czapek's solution agar with cane sugar, pale green or olive, with tough felted mycelium, spreading with broad thin margin, surface growth velvety, consisting of conidiophores only at the margin, but with some tufts of aerial hyphae in center of old colonies; reverse and agar orange to orange red; conidiophores 100 to  $250\mu$  long, with walls rough or granular, 2 to  $3\mu$  in diameter swelling at apex to 5 to  $7\mu$ ; penicilli as single crowded verticils of sterigmata 7 to 10 by  $2.5\mu$ , bearing chains of conidia in a loose column which may become 20 to 30 by  $500\mu$ . Conidia elliptical to globose at times 2 to  $2.5$  by  $2.5$  to  $3\mu$ , quite variable in size, persisting in chains in alcoholic mounts. Gelatine with or without lactose liquefied fairly rapidly, with numerous rhombic crystals in the liquid, with acid reaction which later becomes alkaline.

Grew somewhat more rapidly in bean extract agar, but with same reactions. Decomposes cane sugar with strong acid production, but continued growth brings a return of alkaline conditions.

No. 61 received from Prof. G. F. Atkinson, 1908, as no. 22612b.

69. *P. jantho-citrinum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 311-313; Col. Pl. IX, Cart 1, Pl. XV, fig. 90. 1923.

Colonies on wort gelatine restricted in growth, more or less velvety in appearance, azonate, at first dark green with margin white 1 to 1.5 mm. broad, then a band of bright bluish green, later gray green, coremia none; reverse from pale yellow with spots of orange to violaceous in age, or persisting yellow through the period of observation; odor none; conidiophores  $2\mu$  in diameter below,  $3\mu$  toward the apex, unbranched, smooth; sterigmata 8 to 15 by 3 to  $4.5\mu$ , mostly 8 to 10 by 3 to  $3.5\mu$ , smooth, in verticils of 4 to 12, in a verticil so crowded that their tips diverge, hence probably with conidial chains diverging; conidia ovate 2 to 3.5 or by 1.6 to 3, occasionally  $3.6\mu$ , occasionally  $5\mu$  in long axis, smooth.

Biourge's type no. 1 is recorded as "dead for a long time;" his observation that it formed a serpentine streak in slants was regarded as typical but failure to recover the organism makes identification by others doubtful.

70. *P. subcinereum* Westling. Arkiv för Botanik 11, pp. 58, 137-139; fig. 41, 80. 1911.

Synonym: *P. citreo-nigrum*, Dierckx-Biourge 1923.

Colonies in prune gelatine, thin, gray-green (C.d.C. 347, 372), then ferruginous, with very narrow white margin; vegetative hyphae delicate, not more than  $3.6\mu$  in diameter; reverse uncolored then reddish to dark brownish red, or even dark violet; gelatine slowly and partly liquefied, and partly colored; odor weak but definite and peculiar; conidiophores arising from submerged hyphae, smooth, short, 30 to  $150\mu$  by 2.6 to  $3.4\mu$ , unbranched, swelling at the apex to 3.6 to  $54\mu$ , and bearing monoverticillate penicilli 30 to  $75\mu$  long; sterigmata 7.5 to 11.5 by 2.3 to  $3\mu$ ; conidia smooth or nearly so, elliptical to oblong, 2.3 to 3 or 3.2 by 2 to 2.4, rarely  $2.8\mu$ , swelling in germination to 4 to  $4.8\mu$ .

Species found upon rotten stems. Cultures grew well at 30 to 31°C. and in malt-extract-gelatine, plum agar and potato; upon nutrient agar with 20 per cent sugar the reverse was dark red. Milk and Maranta starch produced poor growth. Westling's culture our no. 2539 grew poorly and was finally lost although his general description was fairly well represented.

71. *P. citreo nigrum* Dierckx, Soc. Scientifique Bruxelles 25: p. 86. 1909. Biourge monogr. La Cellule 33: fasc. 1, pp. 273-274; Col. Pl. IX, Cart. 146, Pl. XV, fig. 87. 1923.

Synonym: *P. subcinereum* Westling, q.v.

Colonies on wort gelatine somewhat spreading; bluish green to sordid rosy; coremia none; reverse at first citrine yellow, then golden, with spots and lines of brown, or almost black; odor, none; conidiophores 15 to 100 $\mu$  or more by 1.5 to 3 $\mu$  figured as arising from creeping hyphae; penicillus figured as a simple verticil of sterigmata or a central axis with terminal verticil of sterigmata and 1 or 2 metulae from the next node, divergent and suggesting monoverticillate stalks of common origin rather than parts of a penicillus; metulae (by typographical error "phialidis") 20 to 30 by 2 to 2.4 $\mu$ , borne as 1 or 2 divergent branches or absent; sterigmata 7 to 13 by 2.5 to 3 $\mu$ , few to many in the verticil; conidia subglobose 1.5 to 3.5 by 1.5 to 3 $\mu$ . Biourge's no. 146 (our no. 4733.35) was apparently received from Amsterdam (Westerdijk) as *P. subcinereum* Westling and later identified with Dierckx's organism from the unpublished colored plates and descriptions. When grown on Czapek's solution agar 4733.35 produced colonies velvety 100 to 200 $\mu$  deep; with unevenly radiating marginal lines growing out into the substratum; bluish gray green; reverse and agar pale yellow with red to brown spots and marginal lines; conidiophores short arising from creeping or trailing aerial hyphae; penicillus monoverticillate with chains parallel or more or less diverging, not in a solid column. Biourge's own statement seems to fix this culture as Westling's type rather than that of Dierckx. In a case of doubt, we prefer to permit the name and description of Westling to take precedence.

72. *P. implicatum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 278-280; Col. Pl. IX, Cart. 76; Pl. XIV, fig. 82. 1923.

Colonies on wort gelatine restricted in growth, much wrinkled, velvety appearing but with close woven hyphae and ropes of hyphae near the substratum; at first sordid grayish blue green with white border, later dark olive green, finally brown; true coremia none; reverse at first white then variegated with rose, bluish, and olive; odor none; conidiophores 30 to 90 by 2 to 3.2 $\mu$ , arising mostly from creeping hyphae and ropes of hyphae; penicillus 10 to 50 $\mu$ , with all walls smooth, figured as mostly a single verticil of sterigmata, occasionally with a branch much lower down on the stalk and divergent (better regarded as a branching hyphae with two short stalked monoverticillate fruits—C. T.); metulae 8 to 25 by 2 to 2.8 $\mu$ ; (when present, practically independent branches.—C. T.); sterigmata 8 to 10.5 by 2 to 3 $\mu$ , in groups of 4 to 8; conidia elliptical to globose, 2 to 3.5 by 1.8 to 2.8 $\mu$ , or, 3 to 4 $\mu$ .

Biourge no. 76 (our no. 4733.73) appears to be type; growth on

Czapek's solution agar, colonies velvety, 100 to 200 $\mu$  deep, radiately wrinkled or buckled, with a narrow outer zone (1 mm.) white, then bluish green, and within dark blue green or green, in age reverse and agar at first colorless, then pale yellow, with purplish or violet and brown shades in older cultures; conidiophores short, arising from submerged or from looping or creeping hyphae, and producing monoverticillate penicilli only, with sterigmata about 8 $\mu$  long and conidia about 3 $\mu$  in diameter; when grown in 15 per cent gelatine in water, velvety appearing with creeping and interwoven hyphae and ropes of hyphae bearing the conidiophores.

Study of Biourge's type leads to the aggregation of several strains not identical with it in details of structure, and reaction but having so much in common and grading into one another to such a degree as to justify grouping them as strains about *P. implicatum*. To prepare species descriptions within this series seems at present impractical. It seems far saner to offer a group or series diagnosis in which something of the range among these forms is indicated and leave to the special worker who may have specific reason, the description of the individual strain in such quantitative terms as will insure its identification.

*P. implicatum* series. Colonies velvety up to 200 to 300 $\mu$  deep, somewhat deeper in center and thinning at the margin; restrictedly growing, with trace of zonation at times in age; blue green to more or less gray shades, reverse in yellow to orange or reddish or maroon shades in age; drops variously colorless, yellowish or sometimes seen as microscopic beads on the hyphae just above the substratum in the marginal areas; conidiophores up to 150 $\mu$  long, by 2 to 3 $\mu$ , usually with apex more or less vesicular (inflated according to Biourge); penicillus with conidial chains in more or less loose or ragged columns, often forming crusts 200 to 300 $\mu$  deep over the surface of the colonies; sterigmata 7 to 10 $\mu$  or even 12 $\mu$  in length in closely packed verticils but rather few in the verticil; conidia 2.5 to 3 by 2 to 2.5 $\mu$  or at times subglobose 2.5 to 3 $\mu$ , smooth.

In addition to Biourge's type, several strains, varieties or species, obviously related have already been examined. Those differ in the intensity of the color of the conidial areas, in the shades of color produced in the substratum and in details of habit.

73. *P. implicatum* var. *aureo-marginatum* n. var. Thom. Type Thom no. 5048.18.

Colonies upon Czapek's solution agar restrictedly growing, velvety up to 200 $\mu$  deep or perhaps 300 $\mu$  in the umbonate center, thinning to a



fimbriate margin about 2 mm. wide, white at first then pale yellow to orange in color with the hyphae studded with microscopic droplets and with the same area marked in reverse by a more deeply orange colored band; conidial areas in blue green shades; reverse in yellow shades passing under some conditions to reddish or flesh color especially in thin layers of substratum; conidiophores up to 100 or 150 $\mu$  long by 2 to 3 $\mu$ , sometimes enlarging upward to 4 $\mu$ , with a vesicular apex up to 6 $\mu$  in diameter; penicilli monoverticillate with conidial chains in loose ragged columns, and forming dense masses or crusts up to 200 to 300 $\mu$  deep in older colonies; which break off in masses when the dish is sharply struck; sterigmata 7 to 10 $\mu$  in close packed verticils, rather few to the verticil; conidia variously 2.5 by 2 $\mu$ , to 3.3 by 2 $\mu$ , or even appearing subglobose 2.5 to 3 $\mu$ .

*Conidia globose: reverse becoming red quickly.*

75. *P. multicolor* Grigorieva-Manilova and Poradielova. Archives des Sciences Biologiques Leningrad 19: 120-134, fig. 1 and plate with photographs 1-6. 1915; in Russian.

Colonies spreading rapidly and broadly zonate, exuding yellow to pinkish or rose drops; gelatine not liquefied; hyphae septate, 2 to 5 $\mu$  in diameter, with pigment grains often showing in the larger hyphae; reverse and substratum dark red with a violet shade in substrata containing sugar, yellow to orange in agar with glycerine; the color changing with the substratum, or entirely absent upon some media; odor slight, moldy; conidiophores as figured, short 1 to 3 celled, usually unbranched, having apex without vesicular swelling, but producing 3 to 5 clavate sterigmata, or less often also producing a divergent branch (or metula?) from the first node with secondary conidial apparatus; conidia smooth but manifestly with wall in 2 layers, globose, 2.2 to 2.5 $\mu$  in diameter, adherent without apparent connective, with long diverging chains, germinating by 1 or 2 tubes.

Species found in Russian soil. The describers placed it in Dierckx's section *Aspergilloides*. Cultures grew readily in the laboratory, producing good growth upon potato, carrot, beet; it did not coagulate milk, but produced a yellow mycelium which became red later. It grew well upon media containing 2 to 3 per cent of lactic acid, but poorly upon alkaline media.

A translation of this paper was furnished by Dr. S. A. Waksman of New Brunswick, New Jersey.

Attempts to obtain the type culture of this species failed. The organism as received either had been replaced by a contaminant or had lost the characteristic appearance it gave when freshly isolated. We isolated at various times from American sources strains suggestive of this description but none of them survived in culture over any long period of time. Notes upon one of these (no. 4236.2 isolated from a rotting mushroom) read as follows: conidial areas gray green with margin orange; reverse in shades varying from cadmium yellow (Ridgway III and II) to flame scarlet; hyphae partly filled with colored substance and often producing color balls at their tips seen as needle crystals in stellate groups in the agar; conidiophores 200 to 300 by 2 to  $2.5\mu$ ; penicilli monoverticillate producing a more or less loose column of conidial chains about  $300\mu$  long; sterigmata 8 to 9 by 2 to  $2.5\mu$ ; conidia 2 to  $2.5\mu$  subglobose.

From its appearance it was called in the laboratory the "paint" organism but either was quickly lost to our collection or under the artificial conditions of culture lost its pigment producing power. A similar form was sent us by Waksman from New Jersey soil but was equally quickly lost.

Another organism in this same series was received among Pribram's collection labeled *A. favedkamp* Kap apparently from Kap Laboratorium in Norway possibly one of Sopp's organisms.

2. *Conidia globose; reverse not in red shades or only very tardily reddish orange.*

*Conidiophore walls rough.*

77. *C. fõlens* Sopp. Monogr., pp. 113-115, Taf. XIII, fig. 96; Taf. XXII, fig. 2. 1912. Suggested as close to *C. coeruleus*, Sopp.

Colonies blue-green, to dark green, becoming brown in old cultures, deep velvety, with heavy wrinkled felt of fine mycelial hyphae; in reverse pale yellow to yellow brown; odor upon all substrata strong, offensive; gelatine only slowly liquefied; conidiophores long, unbranched with walls rough, uniform in diameter with apex broad and flattened; sterigmata not described; conidia about  $4\mu$  in diameter; perithecia not reported.

Species found in soil in Norway. It grew between  $+1^{\circ}$  and  $+35^{\circ}\text{C}$ . not at higher temperatures, produced good colonies in meat-peptone gelatine, in milk, in urine, in meat-peptone broth, in colostrum, in rice and bread, but poorer colonies on potato.

*Conidiophore walls smooth or if pitted faintly and doubtfully so*

This subheading brings together many strains some of them having sufficient affinities elsewhere to be arbitrarily removed. As an example, several organisms velvety spreading, with conidia in columns have the aspect of the *P. Pfefferianum* series no. 24 rather than the *P. glabrum* series hence have been placed in the section already described in spite of the difficulties of identification involved. To remedy this as far as possible a brief sub-key is introduced here.

- a'. Conidial chains in loose columns ..... See no. 24
- a. Conidial chains in compact columns.....b
- b. Restricted or slow growing.....c
- bb. Spreading broadly.....d
- c. Restrictedly growing
- c1. Reverse colorless
  - 1. Forming a very slowly and thinly growing colony with few and scattered columnar masses of green conidia  
*P. columnare* Thom, no. 78.
  - 2. Small, pale gray to olive green colonies reverse uncolored  
4967.7 Melin *P. spinulosum* var. no. 25.
- d. Conidial chains in columns; reverse of colonies mostly in yellow to orange shades differing in details such as mass, shades of color, size and markings of parts.....*P. glabrum*

78. *P. columnare* Thom, n. sp. Type no. 4924 A, from soil at Arlington Farm, Virginia; quickly lost to the collection.

Included because encountered often enough in one summer to convince us that the description covers a strain or series of strains not otherwise accounted for in this book.

Colonies upon Czapek's solution agar, velvety in appearance, with scattered trailing hyphae bearing some conidiophores slowly and very thinly growing, forming solid deep dark green conidial masses in the center of the colony only, with outer areas mostly submerged with scattered zones of columnar conidial masses giving a faintly zonate appearance especially in thin areas of the agar; rhombic crystals evident in agar; reverse uncolored; odor faint; no drops seen; penicillus a solid column about  $20\mu$  in diameter and up to  $100-150\mu$  long suggestive of the appearance of *Aspergillus fumigatus* but lacking the foot-cell of *Aspergillus*; conidiophores  $20$  to  $50\mu$  long by  $2$  to  $2.5$  or  $3\mu$  arising from hyphae up to  $3$  or  $4\mu$  and mostly submerged—some trailing along the surface; all cells rich in protoplasmic contents in both conidiophore and hypha; apex vesicle-like up to  $5$  to  $6\mu$  in diameter with sterigmata from its

whole surface; sterigmata up to 8 to 10 by 2 to  $2.5\mu$ , incurved to form a fairly compact head about 15 to  $20\mu$  in diameter with chains long and adherent into a column which breaks up quickly in alcoholic mount; conidia about  $3\mu$  very faintly punctulate or possibly spinulose, by ordinary examination smooth.

79. *P. glabrum* series.

Study of soil and soil contaminated substances from widely separated sources introduces at this point a great series of species, varieties, races or strains with certain essential characters largely in common and in which large numbers of cultures have been seen and very many of them

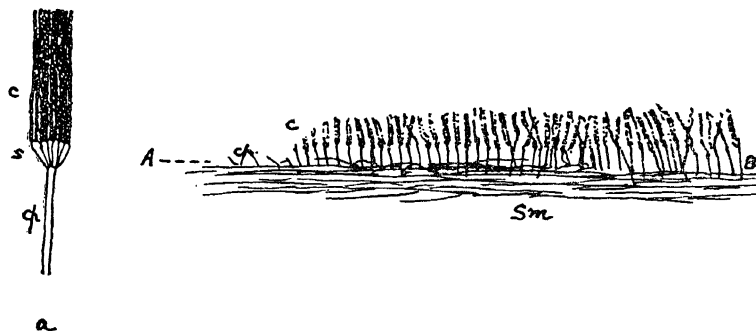


FIG. 26. *P. glabrum* series: a, conidiophore producing a single columnar mass of conidial chains; b, diagrammatic radial section (magnified 25 times) of velvety plane colony showing conidial areas composed of crowded columns of chains, vertical or variously crossed or intertwined. Members of series differ in details of arrangement, measurements, and reactions.

studied. Members of this series from different and widely scattered workers indicate the widespread belief that Wehmer's *Citromyces glaber* belongs here. The general characters include: colonies velvety, usually broadly spreading, some shade of deep green to dark green, mostly becoming brown in age; reverse in a few forms colorless, mostly shades of yellow to orange, in extreme cases becoming brownish orange or red orange even almost red in age; conidiophores erect, unbranched or only occasionally 1-branched near the apex; conidial chains in columns becoming very long; conidia mostly 3 to  $4\mu$ , apparently smooth in some strains, minutely granular or spinulose in others without other distinguishing marks (fig. 26).

A culture brought from Wehmer's laboratory belongs with this series. Subsequent collections show the same general type of organism to be present in Sweden, England, South Africa, and in many parts of the United States. Actual collections examined cover a range from Connecticut to Florida. The cosmopolitan character of the series lead to a deliberate search among Bainier's species for a form which might have represented this type. *P. aspergilliforme* might have been one of them as is suggested by Westling, but no organism of this series was found in the Bainier collection and identification is not possible from Bainier's description on account of his vague statement of great variation in conidial diameter. Similarly, identifications of Sopp's species are too uncertain to justify applying the name of one of them to the series.

Westling's discussion of his species which we have in culture is given first, followed by a diagnostic discussion of one of the commonest organisms in American soil, from which other members of the series diverge in shade of color, length of conidiophore, smoothness or roughness of conidia, and quantitative biochemical characters.

80. *P. frequentans* Westling. Arkiv för Botanik 11, pp. 58, 133-134, fig. 39, 78. 1911.

In Biourge Monogr. La Cellule 33: fasc. 1, pp. 292-293; Col. Pl. X, Cart. 127; Pl. XVII, fig. 99. 1923.

Colonies in prune gelatine velvety in appearance or nearly so, with white margin broad, then areas blue-green (C.d.C. 367, 397, 363, 368) then green (C.d.C. 305, 319, 313) olive green, 289) and finally dark brown; vegetative hyphae 2 to 5 rarely  $8\mu$  in diameter; reverse yellow to reddish yellow; gelatine slowly liquefied, to a yellow fluid with an acid reaction to litmus; conidiophores arising from creeping hyphae with walls smooth, unbranched, from fairly short 60 to 225 up to 600 by 2 to  $3.2\mu$ , with apex swollen to about twice the diameter of the stalk, and bearing a single verticil of sterigmata; penicillus 45 to  $115\mu$  long, consisting of sterigmata and chains of conidia packed into a solid column; sterigmata 8 to 11.5 by 2.2 to  $3.2\mu$ , numerous in a crowded verticil; conidia globose, smooth or nearly so, 2.6 to  $3\mu$  diameter, swelling in germination to 6 to  $7.5\mu$ .

Species common. Type culture our no. 2548 received from Westling; Westling suggests possible identity with *P. aspergilliforme* Bainier; he reports it as producing oxalic acid and another acid that is not citric; his cultures grew freely on all media.

Westling's type is identical with many cultures isolated by us in America.

Biourge's no. 127 (our no. 4733.63) came to us as a form close to if not identical with *P. atramentosum* Thom; his description is however, correct for *P. frequentans*.

Our no. 2467 represents a common form found in American soil. Colonies in Czapek's solution agar with cane sugar, persistently in shades of blue green to green; surface growth of short crowded conidiophores and closely woven trailing or looping hyphae spreading, with very narrow white border in young colonies and some zonation in older colonies; reverse in shades of yellow, orange-yellow to brown with agar in lighter shades of the same color as the mycelium; conidiophores 2 to  $3\mu$  in diameter, swelling at apex to 3.5 to  $5\mu$ , and 100 to  $125\mu$  long; penicillus a single dense verticil of few or many sterigmata cells 7 to 10 by 2 to  $3\mu$  with chains of conidia adhering in a solid column often  $500\mu$  long and 15 to  $20\mu$  in diameter; conidia becoming nearly globose 2.5 to  $3\mu$  and even to  $3.5\mu$  in diameter, granular within, in some races smooth, in others definitely rough, (note—do both conditions appear in same race?) gelatin in water liquefied, not colored, or only slightly brown, with colonies weak pale green; with the addition of cane sugar the gelatin was liquefied much more slowly (not liquefied in ten days) and became a rich brown color; media containing cane sugar became strongly acid by the action of this species. Milk produced growth slowly with small fruiting areas upon the walls of the test tube without discoloration or evidence of digestion in the first eight days.

At one month in one experiment, gelatin in water liquefied by this species required normal sodium hydroxide at the rate of 110 cc. per liter to neutralize the acid present.

81. *C. albicans* Sopp. Monogr, pp. 128–129, Taf. XIV, fig. 101; Taf. XXII, fig. 10. 1912.

Colonies on meat-peptone, sugar gelatine with white mycelium and velvety blue-gray (glaucus) conidial areas, strongly liquefying the gelatine without trace of color; characterized in older cultures by white masses of mycelium overgrowing the conidial areas; odor characteristic, somewhat cheesy; conidiophores long, coarse, septate, clavate toward the apex; sterigmata numerous, in the figure close packed and producing a column of conidial chains; conidia globose, smooth 3 to  $4\mu$  in diameter; perithecia not found.

Species found in cellar-earth in east Norway; colonies grew best at

"room" temperature, slowly at 1°C., grew well in agar and gelatine media, in milk, in acid broth, in wort, on potato, on rice, and on tannin solution. Spores remained viable about 3 years in the laboratory.

Sopp's figure and description would cover any member of this whole series of organisms except for the overgrowths of white mycelium which he uses to characterize the species. No one has reported it since Sopp's work was published, and we are not satisfied to identify any form we have seen by this description.

82. *P. aurantio-brunneum* Dierckx, Soc. Scientif. Bruxelles 25: 86. 1901; see also, Biourge. La Cellule 33: fasc. 1, pp. 309-311; Col. Pl. IX, Cart. 145; Pl. XV, fig. 85; 1923.

Colonies on wort gelatine rapidly spreading, somewhat wrinkled; variously gray green, blue green, to dark olive green, or olive fuscous; coremia none; reverse yellow verging toward reddish, or orange brown; odor none or tardily somewhat ammoniacal; conidiophores unbranched about 50 by 2 to 3 $\mu$ , with all walls smooth; sterigmata 9 to 16 by 3 $\mu$ ; conidia described as globose (rotundis) but given as 3.8 (to 5.5) by 2.5 $\mu$ , but subglobose 3 $\mu$  in our transfers, smooth.

Biourge's type no. 145 (our no. 4733.5) grown upon Czapek's solution agar produced colonies velvety, 100 to 200 $\mu$  deep, with a thin irregular margin fruiting in lines or striae toward the edge, with center wrinkled and more or less tuberculate with tuft-like overgrowth, reverse and agar yellow to deep rich brown, conidiophores about 3 $\mu$  in diameter, with apical verticil of sterigmata 8 to 10 by 2 to 2.5 $\mu$  producing conidial chains massed into 1 solid column 10 to 15 $\mu$  in diameter at base and very long, conidia about 3 $\mu$ , or 2.5 to 3 or even 3.5 $\mu$ .

In our experience this description covers an exceedingly common type of soil organism.

*P. aurantio-brunneum* differs sufficiently from *P. frequentans* to be fairly readily separated. Among the cultures assigned here was 4742P7 from Nottingham, England, 4725.727 collected by D. H. Linder in British Guiana, C. 6885 from Italian bottled water, 4601.B12 on *Vaccinium* from Maine, Cooley's 778 which caused rotting of apples.

83. *P. candido-fulvum* Dierckx. In Biourge, Monogr. La Cellule 33: fasc. 1, pp. 275-277; Col. Pl. X, Cart. 46; Pl. XVII, fig. 98. 1923.

Colonies on wort gelatine, velvety, wrinkled deeply, grayish blue green C.d.C. 367) then dark gray green, at length brown; with marginal

band 1 mm. pure white, 1 mm. translucent; reverse at first almost colorless, then orange, to fulvous or brown; gelatine liquefied; odor none; conidiophores up to 100 by 2 to 2.5 $\mu$ ; penicillus 15 to 25 $\mu$  long with all walls smooth, figured as main axis with monoverticillate apex and occasionally 1 divergent branch commonly longer than the prolongation of the axis; metulae in pairs or none, 7 to 15 by 2 to 2.5 $\mu$ ; sterigmata 9 to 17 by 2 to 3.5 $\mu$ , in groups of 3 to 9; conidia mostly globose 3 to 4.5 $\mu$ , terminal and subterminal spores of the chain 5.5 to 6 $\mu$ .

Biourge's Type no. 46 (our 4733.25) is reported as showing the "phenomenon of Corda" as reported in *P. fieberi*, with the terminal cell of the spore-chain much enlarged. When grown on Czapek's solution agar, the type strain produced colonies spreading broadly, velvety about 400 $\mu$ , sometimes 500 to 600 $\mu$  deep, indistinctly zonate and umbonate in the central areas, with broad thin margin, dull green, in age fulvous or umbrinus; in reverse at first uncolored, then yellowish, with marginal areas brown in age; penicillus either a simple verticil of sterigmata or 1 or more branches 20 to 26 $\mu$  long (equal or unequal in the same group), and chains of conidia massed more or less completely into very long columns; sterigmata mostly 8 to 10 $\mu$  less often up to 18 $\mu$  long in the same verticil; conidia mostly 3 to 3.5, up to 4 $\mu$  in diameter.

84. "*P. citrinum* Thom." In Biourge, Monogr. La Cellule 33: fasc. 1, pp. 295-296; Col. Pl. VII, Cart. 170; Pl. XI, fig. 65. 1923 (not Thom's organism).

Colonies in wort gelatine, restricted in growth or slowly spreading; blue green to green then gray to dark reddish brown; coremia none; reverse sordid yellow, to greenish yellow shades; odor none; conidiophores 3 to 3.2 $\mu$  in diameter, with all walls smooth, arising from creeping hyphae; penicillus a simple verticil of sterigmata about 10 $\mu$  long or with sterigmata or metulae from one or more lower nodes, making a total of 20 to 40 $\mu$  in length; metulae 10 to 16 by 2.2 to 2.8 $\mu$  irregular in origin and number; sterigmata 6 to 9 by 2.5 to 3 $\mu$  in threes or fours in terminal verticils or laterally 1 to many; conidia subglobose, more or less apiculate 2 to 3 at times 4 $\mu$ , swelling to 5 $\mu$  in germinating.

Biourge's no. 170 (our no. 4733.39) is not *P. citrinum* Thom. Grown on Czapek's solution agar 4733.39 produced colonies broadspreading, velvety, dark green, 200 to 300 $\mu$  deep, with margin of radiating uneven lines of hyphae; reverse yellow to orange brown; penicilli developing as columnar masses of conidia, occasionally showing a branch below the apex of the conidiophore producing a secondary column of conidia.



This organism is not *P. citrinum* Thom but a strain or variety of the *P. frequentans* series, not adequately separable as a species.

85. *P. geophilum* Oudemans. Arch. Neerlandaises, 1902, p. 288; Tab. XXV, fig. 1-5.

Colonies on soil extract media round, with alternate zones white and gray green; vegetative hyphae 4 to 8 $\mu$  in diameter; conidiophores about 360 by 6 $\mu$ , hyaline, sparsely septate; penicillus a single verticil of sterigmata; sterigmata about 30 $\mu$  long, commonly 9 in number, lageniform; conidia globose, 3 to 4 $\mu$  in diameter, almost colorless (quasi hyalin), produced in long chains packed into columnar masses.

The striking characters in Oudemans' description were—sterigmata about 30 $\mu$  long, zonation, conidia in columns, 3-4 $\mu$  diameter, coarse stalks. No form with these measurements has been seen by us.

86. *P. glabrum* (Wehmer) Westling, Arkiv för Botanik, 2: no. 1. pp. 131-132; 1911. fig. 77; 1911; change of name based upon description and study of a culture from the "Centrallstelle."

*Citromyces glaber* Wehmer, Beitr. z. Kennt. einh., Pilze I, p. 24; Taf. I, fig. 14-24. 1893.

Colonies with surface growth closely woven, smooth (velvety?—C. T.) not woolly or downy, becoming quickly green to dark green, producing conidia in a heavy masses (literally translated "several mm. high upon the surface"); reverse brownish to dark brown in age, often cracked and peeling off! (reported strongly yellow, dark yellow to reddish yellow by Westling), with yellow coloring matter produced in cooked rice; conidiophore with vesicle-like apex up to 15 $\mu$  in diameter; noted by Wehmer as morphologically identical in its conidial apparatus with *C. pfefferianus* hence with conidiophores up to 70 by 3 $\mu$ ; one series of sterigmata 9 to 14 by 2 to 4 $\mu$  (reported by Westling as numerous) and conidia 2.5 to 3 $\mu$ ; colonies upon media containing high sugar concentrations much wrinkled and folded; grew between 8 and 32°C. with optimum at 20 to 25°C. It produced more acid than *C. pfefferianus*.

No sclerotia or perithecia were observed. Our description follows Wehmer except for additions from Westling's observations as indicated. Several cultures received under the name of Wehmer's species have been members of the *P. frequentans* group but identity with Wehmer's organism has not been established. Dr. Westerdijk contributed two cultures which had come to her collection labeled *C. pfefferianum* (4876.12) and *C. glaber* (4876.11). She could find no differences between

them neither could we. Wehmer in personal conference many years ago declined to identify these species with certainty to individual strains.

87. *P. Oledzkii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 499, 500, 501; Taf. 59; Zaleski no. 626.

Colonies on neutral Raulin with 10 per cent gelatine fairly quickly growing becoming 40 to 45 mm. in diameter in twelve days, liquefying the gelatine quickly and completely, velvety at times zonate in the outer area, central area somewhat convex and more or less wrinkled mostly in a radiate manner; white marginal band 1 to 1.5 mm. wide; in color conidial areas near the margin bluish green then green such shades as C.d.C. nos. 342, 338, 343, becoming dark gray shades of orange yellow such as 168, 173 in old cultures; reverse in yellow to orange shades such as 211, 191, 181; odor none; conidiophores 50 to 150 $\mu$  or 200 $\mu$  by 2.5 to 3.5 $\mu$ , commonly inflated at the apex, figured as arising mostly as branches from trailing aerial hyphae, straight or flexuous, commonly enlarging more or less from base toward apex; sterigmata about 11 to 12 by 2.5 to 3 $\mu$ , mostly in verticils of 5, 8 to 12 or 15, in compact verticils; conidia about 2.5 to 3.5 $\mu$ , smooth, globose or subglobose, showing rather long connectives in the newly formed chains.

Habitat: Species isolated from earth under conifers in the Tatry Mountains in Poland.

Zaleski notes that Biourge placed *P. Oledzkii* near *P. aurantiobrunneum* Dierckx, and *P. frequentans* Westling among the "Aspergilloides Wehmer-Dierckx Series 4: D. Stipes acrocarp. non-divisus Biourge." Our own notes follow: Type strain growing better at 30°C., but well also at 20°C.; colonies upon Czapek's solution agar spreading rather rapidly, velvety plane and zonate in age in outer areas, irregularly wrinkled toward the center, with margin fimbriate, deep dark green (artemisia green to lily green Ridgway LI.), becoming mouse gray in age; reverse ochraceous orange; conidiophores about 100 by 2 $\mu$ ; penicilli single verticils of sterigmata with conidial chains forming columns up to 300 to 400 $\mu$  long by 10 to 15 $\mu$  in diameter; sterigmata 8 to 9 by 2 $\mu$ ; conidia elliptical to subglobose or globose 3 by 2, 2.5 or 3 $\mu$ , smooth.

Culture no. 5010.32 received as type from Baarn in July, 1928, appears to be identified correctly. It belongs to the great group of strains related to *P. frequentans* Westling and cosmopolitan in distribution.

88. *C. robustus* Sopp. Monogr., pp. 125-126, Taf. XV, fig. 103; Taf. XXII, fig. 8. 1912.

Colonies difficult to cultivate, on meat-peptone-sugar-gelatine, velvety,

bristly, gray green, becoming mouse gray in old cultures; in reverse yellow; vegetative hyphae fine, slender, colorless, little branched, few septate forming a rather thin mass enmeshing many crystals of calcium citrate: conidiophores rather coarse figured as enlarging upward like the stalk of *Aspergillus*, unbranched (figured as slightly marked or roughened or studded with crystals as shown in figure but not described), with apex moderately vesiculose; sterigmata coarse, commonly few; conidia smooth, globose  $3\mu$ ; perithecia not found.

Species found in earth in east Norway; colonies grew and produced green fruit at  $1^{\circ}\text{C}.$ , continued to grow up to  $40^{\circ}\text{C}.$ , with their optimum, however, at  $25^{\circ}$  to  $30^{\circ}\text{C}.$  While difficult to grow in meat-peptone media, this species grew well in milk, in urine, in acid broth, on potato, and on rice with little taste or odor.

The species is known only in Sopp's description.

89. *P. sublateralitium* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 315-317; Col. Pl. X, Cart. 57; Pl. XVI, fig. 92. 1923.

Colonies on wort gelatine restricted in growth, more or less wrinkled, velvety, bright bluish green, sometimes truly blue green, then dark blue green, finally fuscous or fuscous gray; coremia none; reverse pale yellowish to pale orange, at length almost brick red; odor weak or none; conidiophores about 70 by  $1.8$  to  $3.2\mu$ , with cell walls smooth, increasing from  $1.8\mu$  at base to the uninflated apex at  $3.2\mu$ , arising from decumbent hyphae, figured mostly as unbranched but with an occasional branch at the topmost node bearing a secondary fruit; sterigmata  $10.5$  to  $15$  by  $2.8$  to  $3.3\mu$  in verticils of 3 to 10; conidia partly ovoid, partly globose,  $2.5$  to  $3.5$  by  $1.8$  to  $2.4\mu$  or  $2.8$  to  $3.2\mu$ .

Biourge's type no. 57 (our no. 4733.119) grown in Czapek's solution agar produced colonies velvety, faintly zonate about  $300\mu$  deep, wrinkled and buckled, with marginal  $500\mu$  white shading to gray green; reverse yellowish to brownish with a tinge of flesh color; agar paler than the mycelium but in the same colors.

No. 4733.119 is probably Biourge's organism, but does not seem to us more than a variety of the *P. frequentans* series of organisms.

90. *P. szulczewskii* Zaleski. In Bul. Acad Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 502, 503, 504; Taf. 60: Zaleski no. 1678.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, fairly rapidly growing becoming 33 to 35 mm. in diameter in twelve

days, liquefying the gelatine quickly and completely, velvety, plane and zonate in the outer area, within showing cerebriform wrinkles, marginal zone white or rosy, 2 to 3 mm. wide in the growing colony; in color, green shades such as C.d.C. nos. 347, 342, 338, 339, 318, 314, with the green fading out in age to dark orange gray shades such as 199, 195; reverse in orange yellow and orange shades such as 153C, 146, 142, 133, 128, 109; odor none; conidiophores 100 to 300 or up to 400 $\mu$  by 2 to 2.5 $\mu$  with apices inflated to 6 to 7 $\mu$ ; penicilli 12 to 15 $\mu$  in length; sterigmata about 9 to 11 $\mu$  by 2.5 to 3 $\mu$ , in compact verticils of as few as 6, more commonly 16 to 24, with short broad tubes; conidia about 2.5 to 3 $\mu$ , smooth, globose, showing a short but definite connective when young.

Habitat: Species isolated from earth under conifers in square "317" of the forest "Puszcza Bialowieska" in Poland.

Zaleski reports that Biourge regarded it as a variety of *P. Terlikowskii*, that further study established differences in the size of conidia, in the average length and diameter of the conidiophores, in the numbers of sterigmata in the verticil and in the form, size and abundance of the ampullae or vesicle-like swellings at the apex of the conidiophores. In colony appearance, color of conidial area and reverse the two species are very similar. He places it among the "*Aspergilloides* Wehmer-Dierckx Series 4: D. Stipes acrocarp: non-divisus Biourge." No culture of this species was received but it is placed by us here from Zaleski's figures and description.

*Conidia rough or spinulose*

91. *P. baiiolum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 305-306; Col. Pl. VIII, Cart. 117; Pl. XIII, fig. 74; 1923.

Colonies in wort gelatine, velvety to sublanose, wrinkled, pale blue green, then grayish blue green, gradually darkening to a reddish fuscous; coremia none; reverse sordid yellow, to yellow brown; odor none; conidiophores 2 to 3 $\mu$  in diameter, increasing gradually from base to apex, with walls smooth, penicillus a simple verticil of sterigmata, occasionally with extra sterigmata lower on the stalk than the vesicular apex; sterigmata 9 to 12 by 3.5 $\mu$  in verticils of 4 to 10; conidia elliptical then nearly globose 2.8 to 4.8 by 2.5 to 4.8 $\mu$ , in germinating 6 by 5.2 $\mu$ , smooth at first, rugose echinulate when ripe.

Biourge type no. 117 (our no. 4733.17) when grown upon Czapek's solution agar produced colonies with margin white, and 1 to 2 mm.

broad, velvety, uneven, about 300 to 400 $\mu$  deep, with some floccose overgrowth and up to 700 $\mu$  deep in older areas, dirty gray green, becoming darker in age; reverse pale yellowish orange with center dark; conidial chains massed into very long columns otherwise our findings are fully in agreement with Biourge.

A culture from Dr. Elias Melin from forest soils (5007.84) comes fairly close to this species.

92. *P. flavi-dorsum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 290–291; Col. Pl. VIII, Cart. 87; Pl. XIII, fig. 73; 1923.

Colonies in wort gelatine wrinkled, velvety with central overgrowth (duveteuse), spreading with broad white margin (4 mm.); blue green toward violet, at length dark fuscous (C.d.C. given as very dark orange yellow shades 164–165); coremia none; reverse from sordid yellow to fulvous; odor none; conidiophores 2.5 to 3 $\mu$  in diameter with walls squamulose! penicillus figured as a simple verticil of sterigmata packed parallel with apparently chains of conidia parallel (or adherent into a column? no observations given—C. T.): metulae rare, long, 30 to 45 by 2.2 to 2.8 $\mu$ ; sterigmata 9 to 14 $\mu$  long, with walls echinulate-squamulose, in verticils of 4 to 10; conidia subglobose 2.5 to 4.4 $\mu$ .

Biourge's type no. 87 (our no. 4733.60) grown in Czapek's solution agar produced colonies azonate, wrinkled, velvety, spreading, 150 to 300 $\mu$  deep, pale bluish green, to dull gray green; reverse colorless (at 7 days), yellow showing spots of reddish brown; sterigmata as in Biourge except an occasional branched sterigma producing a secondary verticil; conidia subglobose 3 $\mu$  rarely more than 3.5 $\mu$ , with a trace of pitting or echinulation of wall.

The appearance of the wall of the sterigmata and conidiophore designated squamulose can be seen by careful use of an oil immersion but hardly fulfills the usual implications of the term. Some markings of the cell-wall are present but only in part of the individual conidiophores.

## CHAPTER XIII

### MONOVERTICILLATA-RAMIGENA

*Section II. Ramigena: Fertile hyphae* (conidiophores?) prostrate or ascending, mostly branching, with branches 1-celled (metula-like), or long and septate, simple or occasionally again branched, each bearing a terminal monoverticillate penicillus, but not producing definite apical verticils of metulae or branchlets (fig. 27).

In these species the individuality of the monoverticillate penicillus is evident but divaricate branching at various levels but without definiteness of organization or arrangement is constantly observed.

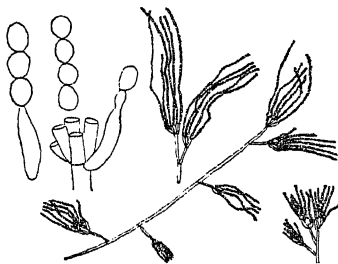


FIG. 27. A ramigenous type of conidiophore and penicilli

This section is arbitrarily created to provide for a group of forms mostly described and figured by Bainier and Sartory as species of *Citromyces* in which the monoverticillate type of penicillus is characteristically produced but the fertile hypha instead of being an erect conidiophore tipped by a single penicillus, is a creeping or ascending fertile hypha bearing branches from several septa varying in appearance from that of simple conidiophores to that of fruiting cells or metulae near the apex of a main axis (fig. 28).

The cultures received from the Bainier collection under these names failed to comply with Bainier's description to such a degree that we are compelled to question whether we have authentic cultures of any of them. In dealing with these species, Bainier's descriptions appear to have been based upon colonies grown upon licorice sticks, hence he automatically fails to give colony characters. Our own collections, however, do show that there are species which produce these branching fertile

hyphae with each branch producing a monoverticillate penicillus. This group of descriptions has, therefore, been kept together in the expectation that some of them may again be recognized.

Certain of Zaleski's species have been added to the Section after we had studied his series of cultures upon our own substrata.

#### Ramigena.

##### Conidia elliptical

Conidia 4 by  $2\mu$ ; reverse and agar yellow orange to red. .*P. cyaneum*, 95.

Conidia 3 by  $2\mu$ .....*C. musae*, 96.

##### Conidia globose

Conidia  $2\mu$ , conidia in diverging chains; reverse but

not agar rose.....*C. minutus*, 97.

*C. cesiae*, 19.

Conidia  $2\mu$ ; branching cymose.....*C. affinis*, 98.

Conidia 2.5 to  $3\mu$ ; white to slowly blue green then green. .*C. brevis*, 99.

Colonies dull gray green, conidia 2.5 to  $3\mu$ .....*P. Waksmani* Zal.,

100

Conidia  $2.8\mu$ ; reverse rose or peach.....*C. ramosus*, 101.

Colonies gray or greenish gray, deeply floccose; reverse

colorless or rosy; conidia 2 to  $2.5\mu$ .....*P. Siemazki* Zal., 102.

Colonies closely felted olive gray; reverse colorless to

rosy.....*P. Jensenii* Zal., no.

241.

Conidia 2 to  $2.5\mu$  or slightly larger, in columns.....*P. Sartoryi*, 103.

95. *P. cyaneum* (B. and S.) Biourge. Monog. List Onomastique, p. 102.

Synonym: *C. cyaneus* Bainier and Sartory. Bul. Soc. Mycol.

France 29: 157-161; Pl. IV, fig. 4. 1913.

Colonies upon licorice sticks forming small colonies with mycelium spreading slowly, floccose or hirsute with rather long erect or ascending, branching hyphae, becoming conspicuously bright blue in color (C.d.C. 392, 397, 398) with conidium formation, with a sterile margin canary yellow; reverse yellow orange to red; conidiophores mostly arising as diverging branches from erect or ascending hyphae each with its branches terminated by an obconical vesical apex with a verticil of 8 to 12 sterigmata, about  $11.2\mu$  in length producing chains of conidia packed into a columnar mass; conidia elliptical about 4 by  $2\mu$ . (Fig. 28 f.)

Species found upon the rind of an orange. Cultures liquefied gelatine coagulated and peptonized milk, and produced citric acid from glucose.

In our series of cultures from the Bainier collection, no. 4640.423 was labeled *C. cyaneus* and 4640.422 *C. cesiae*. The culture labeled *C. cyaneus* had conidia globose, smooth, 2 to  $2.5\mu$  in diameter which excludes it from Bainier and Sartory's species. However, 4640.422 was found to

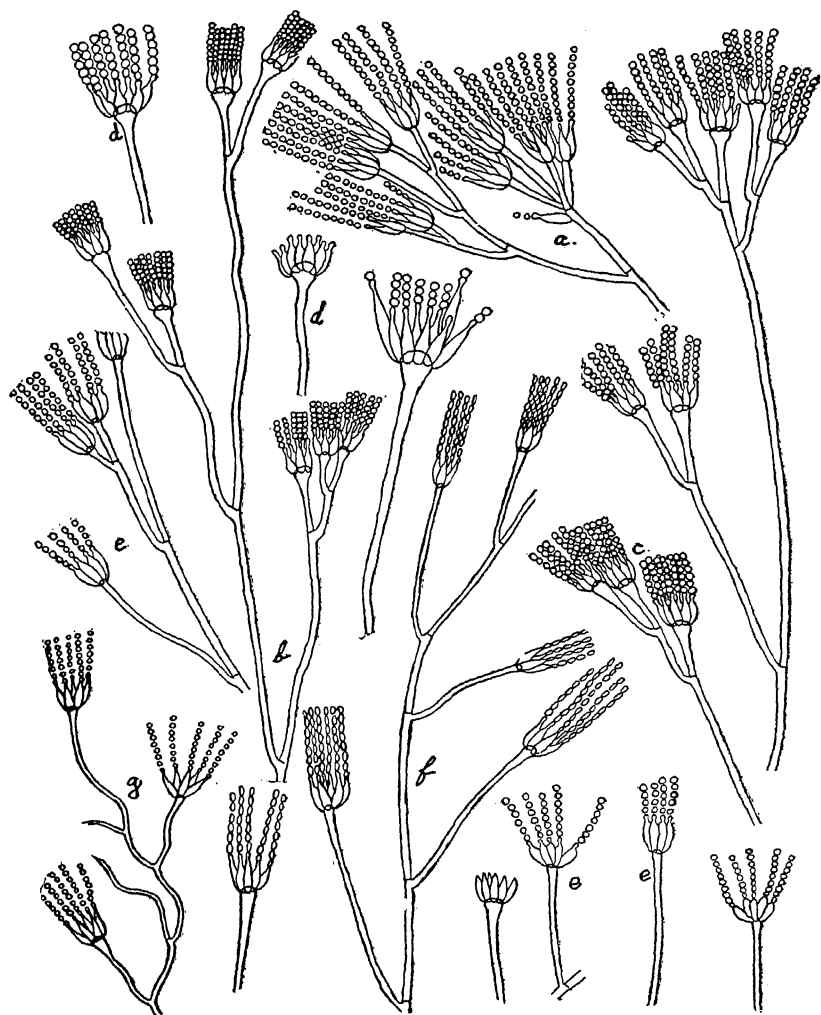


FIG. 28. Ramigenous species from Bainier and Sartory: a, *C. ramosus*; b, *C. affinis*; c, *C. subtile*; d, *C. brevis*; e, *C. cesiae*; f, *P. cyaneum*; g, *C. minutus*.



reproduce this description of *C. cyaneus* closely enough to suggest that it probably is the type culture of *C. cyaneus*. Our notes follow: Colonies in Czapek's solution agar forming a close felt with almost velvety appearance, at times with a fairly deep floccosity at the surface of the substratum; with mycelium consisting of hyphae 1 to  $2\mu$  in diameter tearing easily; varying in color from pale dull glaucous blue near the margin to deep bluish gray green in center (Ridgway XLII.41''); with reverse colorless or in slanted tubes yellow in the deep areas, rosy in the shallower and drier areas; becoming rosy throughout in age; conidiophores 100 to 300 by  $2\mu$ , rising separately from the substratum or as branches from ascending aerial hyphae; penicilli monoverticillate with narrow columnar masses of conidia; sterigmata varying up to 15 or even  $20\mu$  in length, few and closely packed in the verticil; conidia 3 to 4 by 2 to  $2.5\mu$ , smooth.

Another strain, no. 5031.137, from the British Cotton Industry Research Association produced colonies in Czapek's solution agar (at about 20 to 25°C.) velvety, azonate, 2 to 3 cm. in diameter in seven days, and about 200 to  $300\mu$  deep, plane, with sterile margin about 1 mm. in width, next a pale blue conidial area about 3 to 4 mm. wide and a central area grayer with pronounced development of cinnamon to reddish drops which fused into large drops in the central area; in reverse mycelium and agar were yellowish through orange to orange red or red shades; the same color extended 2 to 3 cm. beyond the colony gradually fading out.

Conidiophores erect or ascending especially at the margin showed a succession of branches each with a monoverticillate penicillus, or rarely groups of divaricate branches but mostly showed the ramigenous character of the section.

Sterigmata were up to 10 by 2.5 to  $3\mu$ ; conidia about 3 to 3.5 by 2 to 2.5 or sometimes almost subglobose  $3\mu$ , smooth or nearly so.

96. *C. musae* Bainier and Sartory. Bul. Soc. Mycol. France 29: 154-157; Pl. V, fig. 1-3. 1913.

Colonies on licorice sticks green (near C.d.C. 318) with interlacing aerial mycelium producing erect hyphae bearing the conidiophores, at intervals as short divergent branches  $2\mu$  in diameter, enlarging at the vesicular apex up to  $8.5\mu$ ; sterigmata 6 to 8 in the verticil mostly on the upper part only of the vesicle, occasionally borne separately lower down 7 to 8 by  $3\mu$ ; conidia elliptical to globose, 3 by  $2\mu$  or globose  $2\mu$  in diameter.

Species found on the skin of a banana. Cultures grew best at 25 to

27°C. but showed a range of growth between 15° and 38°C. They liquefied gelatine, coagulated and peptonized milk and produced citric acid from glucose.

The type culture has not been seen and thus far we have not been able to refer any strain to this species. In transferring the species of *Citromyces* to *Penicillium C. musae* of Bainier and Sartory would be invalidated by *P. musae* Weidemann 1907. No change is offered since no material is referred by us so far to this species.

97. *C. minutus* Bainier and Sartory. Bul. Soc. Mycol. France 29: 137-144; Pl. IV, fig. 3. 1913.

Colonies upon licorice sticks cottony floccose up to 5 mm. or more deep, long white, then developing a delicate tint of gray green (C.d.C. 346) which becomes deeper in age, but finally becomes rose; aerial mycelium much branched, undulate and terminating in sterile tips; reverse of mycelium rose upon several media but no pigment excreted; conidiophores slightly sinuate arising as short branches at intervals upon sinuate aerial hyphae, with apex slightly dilated, each bearing a verticil of 5 to 6 sterigmata, about  $8.4\mu$  in length, with tips diverging; conidia globose up to  $2\mu$  in diameter, in diverging chains. (Fig. 28 g.)

Cultures grew within a range of 18° and 40°C., with the optimum about 26 to 28°C. It grew well upon the usual media tested, liquefied gelatine in 9 to 10 days, coagulated and peptonized milk, and produced citric acid from glucose.

Their type of this species has not been seen by us. From the figures it might be placed here or with the more strictly monoverticillate series near *P. decumbens* or *P. citreo-viride*. No change of combination is offered since no culture is referred to this species.

No. 4733.84 labeled *P. luteo-viride* Biourge received as Biourge no. 98 shows the morphology of *C. minutus* B. & S., but differs in the colors produced in the substratum from the colors reported for *C. minutus*.

98. *C. affinis* Bainier and Sartory. Bul. Soc. Mycol. France 28, fasc. 1, pp. 39-43; Pl. I, fig. 1-7. 1912.

Colonies on licorice sticks spreading broadly, forming a thick floccose heavy growth, at first white then green, finally sordid dark green from the great quantities of conidia produced; conidiophores about  $2\mu$  in diameter, smooth, produced as branches and systems of branches from aerial hyphae, figured as sinuate, each commonly tipped with a verticil of sterigmata then the terminal segment equalled or surpassed by a

secondary, diverging branch produced at the next node below, bearing second conidial mass, and the process repeated often 2 to 3 times conidia about  $2\mu$  in diameter. (Fig. 28 b.)

Species found upon a willow basket. Cultures grew best at  $24^{\circ}$  to  $26^{\circ}$ , and with a range from  $18^{\circ}$  to  $40^{\circ}\text{C}$ . with richest growth on potato and glycerine. Colonies grew well upon all common media, liquefied gelatine, coagulated milk, digested the curd slowly, produced citric acid from glucose.

We have not identified this species.

99. *C. brevis* Bainier and Sartory. Bul. Soc. Mycol. France 28, fasc. 1: 43-45, Pl. II, fig. 1-4. 1912. (Fig. 28 d.)

Colonies growing well on various media forming a thin felt of aerial mycelium, white then slowly (three to seven days) blue green (variously C.d.C 367, 378, 396 and related), becoming green (343) in old cultures in some media; conidiophores arising as branches of trailing or ascending hyphae, 2 to  $2.5\mu$  in diameter, and varying considerably in length, somewhat sinuous, with walls smooth, swelling at the apex to form a vesicle 7 to  $8\mu$  in diameter; sterigmata up to  $10\mu$  in length, figured as packed closely parallel the verticil; conidia globose, 2.5 to  $3\mu$ , figured as forming closely parallel (? adherent) chains; perithecia not reported.

Optimum temperature for culture  $26$  to  $28^{\circ}\text{C}$ . Cultures liquefy gelatine, coagulate milk and peptonize the casein, and produce small amounts of citric acid in glucose media.

Culture no. 4640, 421 received in the Bainier collection was not a member of this group at all. We have not identified a strain as *C. brevis*.

100. *P. waksmani* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 468, 469; Taf. 49; Zaleski no. 1554.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 28 to 3.3 cm. in twelve days, with the gelatine slowly but completely liquefied, in surface velvety, occasionally showing zonation in the outer areas, with regularly radiate wrinkles 2 to 4 mm. wide and 1 to 2 mm. in height, and central area elevated and more or less overgrown with secondary mycelium; margin white 1 mm. in width during the growing period; in color conidial areas blue green C.d.C. in such shades as 378B, 396, 347, 367, 366, becoming dark orange brown colors such as 139, 140 in old colonies; reverse and liquid showing more or less definite zones, pale yellow to orange in such shades as 146, 171, 131 136, 137; odor faint, agreeable; conidiophores varying in length  $100\mu$ ,

more commonly 200 to 300 $\mu$  by 2.5 to 3.5 $\mu$ , straight or flexuous, simple or sparingly branched, with apices vesicle-like, with walls smooth; penicillus 20 to 28 $\mu$  in length; metulae varying in length in the verticil 10 to 16 or even 18 $\mu$  by 3 to 4 $\mu$  in groups of about 3 to 6, often increasing in diameter from base to apex and diluted at the apex; sterigmata about 8.5 to 10 by 2 to 2.5 $\mu$ , in verticils of about 6 to 12, straight, fusiform, showing mostly at slender tube; conidia 2.2 to 2.5 $\mu$  (up to 2.8 $\mu$ ), smooth, globose, showing connectives.

Habitat: Species isolated from earth under mixed woods in square "369" in the forest "Puszcza Bialowieska" in Poland. Zaleski notes that Biourge regarded this as related to *P. chrysogenum* Dierckx (?CT) nevertheless he places it in "Biverticillium Dierckx subsec. 3. Radiate-undulata." Our notes follow: Type strain growing well about 20°C., poorly at about 30°C.; colonies on Czapek's solution agar at 20°C., azonate, slowly spreading, velvety appearing but with a close woven basal felt of fine aerial hyphae bearing conidiophores as ascending criss-crossing rather than erect branches, with central area somewhat elevated, outer areas plane and a narrow white (1 mm.) margin, dull gray green; reverse colorless; drops not seen; odor, none detected; conidiophores inflated at apex, very short to 100 $\mu$  or even 200 $\mu$  long by about 2 $\mu$  in diameter, or developed as long trailing hyphae; penicilli partly single verticils of sterigmata, partly showing one or more branches with secondary penicilli, from the topmost septum, commonly exceeding in length the continuation of the main axis; sterigmata 9 to 11 by 2 $\mu$ , diverging at the tips; conidia 2.5, or scarcely 3 $\mu$ , more or less globose with walls faintly pitted or irregular.

Culture no. 5010.36 received as type from Baarn in July, 1928, fits Zaleski's descriptive terms better than his figures. In our cultures it appears to be closer to Bainier's *Citromyces ramosus* than to the Radiata of Biourge (*P. chrysogenum* series).

101. *C. ramosus* Bainier and Sartory. Bul. Soc. Mycol. France 29: 144-148, Pl. IV, fig. 1 and 2. 1913.

Colonies on licorice sticks green (C.d.C. 268) showing a trace of bluish at first upon some media, with margin usually rose or peach in color, in age almost violaceous; mycelium rather thin, spreading slowly, consisting of creeping hyphae much branched and interwoven with dense conidial masses making the details of structure difficult to determine; reverse showing rose or peach color, but not conspicuous ("peu apparent") conidiophores figured as complex systems of diverging branches,

separate or in verticils, terminal branches or metulae short enlarging upward to produce verticils of few to a dozen sterigmata about  $8\mu$  in length, with chains of conidia figured as parallel; conidia globose about  $2.8\mu$  in diameter. (Fig. 28 a.)

Species associated with *C. cesia* in the insect tunnels, cultures grew best at  $26^{\circ}$  to  $27^{\circ}\text{C.}$ , liquefied gelatine slowly (in 17 days), coagulated and peptonized milk, produced small amounts of citric acid from glucose.

*Citromyces ramosus* Sopp is listed by Biourge (Monogr., p. 105) but no reference to the name is found in Sopp's book.

102. *P. Siemaszki* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 487, 488; Taf. 54; Zaleski no. 1696.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 25 to 30 mm. in diameter in 12 days, liquefying the gelatine quickly and completely, velvety or slightly floccose, plane and zonate only in the outer area, central area more or less depressed and thrown into broad wrinkles; white margin about 2 mm. wide in the growing colony; in color pale blue green 378B when young, later 372, 368, to green 347, 348, and in age orange brown shades such as 173 and 148; reverse and liquefied gelatine in orange-yellow shades such as 171, 156, 161, 128; odor strong, suggesting potatoes rotted by fungi; conidiophores 100 to 300, or even to 500, by  $2.5$  to  $3\mu$ , usually flexuous, simple or sparingly branched, with apices inflated and with all walls smooth; penicilli 10 to  $12\mu$  long when simple, 20 to  $28\mu$  long when branched; metulae 10, 12 to 16 or even to  $18\mu$  by  $2.5$  to  $3\mu$ , commonly 2 or 3 in the group and unequal in length; sterigmata about 9 to 10 by  $2.3$  to  $2.8\mu$ , commonly in verticils of 5 to 12 or 15, with short tubes; conidia  $2.3$  to  $2.5$  or even to  $3\mu$ , smooth, globose, showing a definite connective.

Habitat: Species isolated from soil in square "317" of the forest "Puszcza Bialowieska" in Poland.

Zaleski reports that Biourge examining his material placed the species with *P. sulphureum* Sopp but Zaleski places it in the Series Corylophilum of the Aspergilloides. Our notes follow: Type strain growing better at  $20^{\circ}$  than at  $30^{\circ}\text{C.}$ ; colonies upon Czapek's solution agar about  $20^{\circ}\text{C.}$ , deeply floccose with very fine ( $2\mu$  in diameter) close woven hyphae, forming a mass 2 to 3 mm. deep in center thinning to a submerged margin 1 to 2 mm. wide, with more or less radiate wrinkling, and a trace of zonation in the fully developed colony; in color gray with irregular zones of greenish and with white overgrowth in central area; reverse colorless or a faint rosy ("peach"); drops, colorless, central; conidiophores about  $2\mu$  in diameter; penicillus either a single verticil of 3, 4, to

several (always few) sterigmata, with long parallel or diverging chains of conidia, or with one or more diverging branches and secondary verticils; sterigmata about  $8\mu$  in length, few in the verticil; conidia elliptical to subglobose 2 to  $2.5\mu$  in diameter, persisting in chains when mounted, with connectives evident.

Culture no. 5010.21 received as type from Baarn in July, 1928, complies closely enough with Zaleski's description to be accepted. In our culture this species differs only in minor detail from Zaleski's *P. jensenii*.

103. *P. Sartoryi* Thom syn. *C. subtilis* Bainier and Sartory. Bul. Soc. Mycol. France 28, fasc.

Synonym: I, p. 46, Pl. II, fig. 5-7. 1912. Not *P. subtile* Berk. no. 613.

Colonies on licorice sticks forming a deep heavy growth, at first white then green (C.d.C. blue green 396, 366) to ashy gray; gelatine not liquefied in 16 to 17 days; conidiophore figured as long with an apical group of diverging branches, some of them almost metula-like, each tipped with a vesicular apex 8 to  $10\mu$  in diameter, globular or obconical, with a verticil of sterigmata and conidial chains parallel (in a column?); sterigmata few to many and unequal in length in the verticil; conidia 2 to  $2.5\mu$  or slightly larger, in columns.

Cultures grew best at  $25^{\circ}$  to  $28^{\circ}\text{C}.$ , failed to produce citric acid from glucose in many experiments, coagulated milk and partially peptonized the curd. It did not grow well on potato alone but grew well when glycerine was added to potato. Carrot produced good growth.

We have had at least six organisms in culture which showed long conidiophores with a terminal group of *Citromyces* fruits on branches of different length and at different levels as described for *C. affinis*. There manifestly exists a related group with this general morphology.

Another culture with this morphology no. 5048.14, grown on Czapek's solution agar produced colonies floccose composed of closely felted hyphae about 2 to  $3\mu$  in diameter, forming colonies about  $300\mu$  deep, slowly and more or less restrictedly growing with margin somewhat fimbriate from radiating submerged hyphae, in color deep dull blue green to bluish gray; reverse colorless or yellowish; conidiophores partly long trailing hyphae 300 to 400 by  $2\mu$ , partly branches of aerial hyphae 100 to  $200\mu$  long, variously branching with the secondary branches often longer than the main axes, sometimes resembling terminal verticils of metulae, each terminating in a monoverticillate penicillus; sterigmata conidia 2 to  $2.5\mu$  less often  $3\mu$  in diameter.

The change of name is necessary to avoid confusion with *P. subtile* Berk.

## CHAPTER XIV

### ASYMMETRICA-VELUTINA

#### *II. Asymmetrica.*

Penicillus consisting of two or more series of elements including sterigmata and metulae with or without branches (rami) of one or more series, with the branching system typically lop-sided, one-sided, or asymmetrical.

The groups of elements in such penicilli develop ordinarily in basipetal succession—the first group of sterigmata upon the tip of the main axis, a verticil of metulae from the next lower node or septum, and lower branches when present successively lower down on the conidiophore and incompletely verticillate or more or less one-sided in arrangement.

This general type of penicillus as described is an arbitrary common character bringing together several more or less natural groups of species and many single species whose affinities are very little known. General colony characters and more or less minor morphological differences are made the basis for sections and subsections.

The following dichotomous key to the sections proposed may aid in locating species.

- I. Aerial hyphae simple or branched but not combined into ropes or fascicles.....II.
- I. Aerial hyphae partly at least combined into ropes, fascicles or coremia.....V.
- II. Colonies velvety (velutinous): including species with surface networks of aerial hyphae but general velvety effects.....*Asymmetrica-Velutina.*
- II. Colonies floccose or lanose: including some species with velvety margins in age.....III.
- III. Colonies floccose or lanose: with well marked development of aerial felts within which the conidial areas develop.....IV.
- III. Colonies floccose, or floccose in the central area with marginal area almost velvety; penicilli with short compact base and diverging sterigmata with conidial chains parallel or diverging.....*Brevi-compacta.*
- IV. Colonies floccose with complex branching penicilli aggregated to form the typical brush or broom.....*Lanata-typica.*





- VIc. Colonies gray rose, conidia 2 to 4 $\mu$  in long axis... *P. exiguum* Bainier  
429.
- X. Colonies some shade of green or olive.....XI.
- XIa. Colonies olive green; conidia 5 to 8 by 4 to 7 $\mu$ ....*P. digitatum* series.  
XII.
- XIb. Colonies dark green; conidia 3.6 to 6.8 by 2.3  
to 3 $\mu$  in deep masses breaking off easily.....*P. oxalicum* series.  
XIII.
- XIc. Colonies gray green; conidia 2 to 5 by 1.5 to 4 $\mu$ ..*P. duponti* G. & M.,  
121.
- XId. Colonies gray green; conidia 5 to 8 by 3.5 to 4.2..*P. lanoso-grisellum*  
Biourge, 115.
- XIe. Colonies blue green; conidia 7 by 5 to 6 $\mu$ .....*P. viridum* Sopp, 678.
- XII. On citrus fruits
1. olive green; conidia 5 to 8 by 4 to 7.....*P. digitatum* Sac-  
cardo, 111.  
Synonym—conidia 5 to 8 by 4 to 7.....*P. olivaceum* Weh-  
mer, 111.
  2. White; conidia 5 to 8 by 4 to 7.....*P. digitatum* Sacc.  
var. *Californicum*  
Thom n. var., 112.
  3. Gray green; conidia 5 to 8 by 4 to 7.....*P. lanoso-grisellum*  
Biourge, 115.
  4. Syn. of *P. digitatum*.....*P. olivaceum* var.  
*italicum* Sopp, 114.
- XII. Colonies as in *P. digitatum*; conidia 4 by 3 $\mu$ ....*P. schneeggii*—no  
330. Chapter  
XIX.
- XIII. On soil, manure, etc.....XIV.
- XIVa. Conidia 3.6 to 6.8 by 2.3 to 3 $\mu$ .....*P. digitatum* var. *dis-*  
*coideum* Marchal,  
116.
- XIVb. Conidia 12 to 18 by 6 to 10 $\mu$ .....*P. olivaceum* var.  
*Norvegicum* Sopp,  
118.
- XIVc. Probably the same form.....*P. Wurceburgense*  
Biourge, 119.
- XIVd. Conidia up to 5 by 3.5 $\mu$ —breaking off in masses..*P. oxalicum* Thom  
and Currie, 120.
- XIVe. On manure, gray green; conidia 2 to 5 by 1.5  
to 4 $\mu$ .....*P. duponti* G. & M.,  
121.
- XIVf. Conidia 7 by 5 to 6; penicillus divaricate.....*P. viridum* Sopp, 678.

XX. Conidia less than  $4.5\mu$  in long axis.....XXI.

XXI. Penicilli with metulae divaricate producing verticils of conidia approximating separate monoverticillate penicilli.....XXII.

XXI. Penicilli more compact in the characteristic form of a brush or broom.....XL.

Sub-section 2. Velutina-divaricata

XXII. Conidia definitely elliptical.....XXIII.

XXII. Conidia when ripe globose or subglobose.....XXV.

XXIII. Conidia about 3 to 4 by 2 to 3 or 3.5..... XXIV.

XXIV. Colonies about  $200\mu$  deep, blue green; reverse yellow to orange.....*P. rubens* Biourge, 125.

XXIV. Closely related form.....*P. rubrum-fide* Biourge, 126.

XXIV. ....*P. obscurum* Biourge, 127.

XXIV. Colonies dull dark green; reverse becoming purple brown or almost black.....*P. atramentosum* Thom, 128.

XXV. Conidia globose or subglobose.....XXVI.

XXVI. Conidial chains more or less divergent and tangled in age.....XXVII.

XXVI. Conidial chains in fairly compact columns....XXXV.

XXVII. Conidia rough 3 to  $3.5\mu$ ; bluish to greenish gray; reverse citrine to orange brown.....*B. glauco-griseum* Sopp, 240.

XXVII. Conidia smooth.....XXVIII.

XXVIII. Colonies restrictedly growing.....XXIX.

XXVIII. Colonies spreading broadly.....XXX.

XXIX. Colonies pale bluish green about  $200\mu$  deep; conidia 3 to  $3.5\mu$ .....*P. fellutanum* Biourge, 52. See Chapter XII.

XXX. Colonies spreading; dull green to gray green; reverse uncolored or slightly purplish in deeper areas; conidia about  $4\mu$ .....*P. chloro-leucon* Biourge, 131.

- XXX. Colonies in outer areas velvety plane, more or less floccose in center, bluish gray green to olive green; conidia 2 to  $2.5\mu$  ( $2.8\mu$  in Zaleski)... *P. Westlingi* Zal., 132.
- XXXV. Conidial chains in fairly compact columns and the columns divergent.....XXXVI.
- XXXVI. Reverse uncolored, greenish to bluish black.... *Corylophilum* series XXXVII.
- XXXVI. Reverse in yellow shades..... *Citrinum* series XXXVIII.
- XXXVII. Colonies dull green; reverse reaching greenish black shades; conidia 2 to 3 or even  $3.5\mu$ ..... *P. corylophilum* Dierckx, 133.
- XXXVII. Colonies gray green; reverse yellow on wort, black on potato; conidia 3 to  $3.5\mu$ ..... *P. umbonatum* Sopp, 134.
- XXXVIII. Reverse in yellow shades.....XXXIX.
- XXXIX. Colonies broadly but indistinctly zonate, thin at margin, up to  $500\mu$  deep in center, grayish blue green—reverse more or less zoned yellow; conidia 2 to  $2.5\mu$ ..... *P. Steckii* Zal., 135.
- XXXIX. Colonies narrowly growing, plane, thin, dull blue green; conidia 2.4 to 3 or even  $3.5\mu$ ..... *P. citrinum* Thom, 136.
- Syn. based upon Biourge's type no. 4733.14.... *P. aurifluum* Biourge, 137.
- XL. Branches and metulae more or less compacted into the brush or broom type of penicillus....XLI.
- XLI. Colonies regular in outline, velvety in appearance, mostly with a network of aerial hyphae at the base of the conidial area; conidiophores mostly ascending rather than erect, directed outward near the margin but extensively criss-crossing throughout the colony; penicilli characterized by terminal verticils of metulae more or less closely aggregated and each subtended by one or more distant secondary divergent branches bearing secondary penicilli often monoverticillate...Radiata XLII.
- XLI. Conidiophores and penicilli more erect, penicilli more completely aggregated into a morphological unit.....L.

## Radiata or P. Chrysogenosum Series

- XLII. Drops usually colorless.....XLIII.  
 XLII. Drops usually yellow.....XLIV.
- XLIIIa. Colonies 300 to 400 $\mu$  deep, blue green to green,  
 to brown; reverse and agar yellow or yellowish;  
 in age conidia from elliptical 4 by 3.3 at first to 4 $\mu$ .....*P. chrysogenum*  
 Thom, 140.
- XLIIIb. Colonies up to 600 $\mu$  deep, blue green to gray  
 to purplish gray; reverse yellow at first then brown;  
 conidia subglobose when ripe 3.6 to 4.2 by 3 to 4 $\mu$ .....*P. chlorophaeum* Bi-  
 ourge, 141.
- XLIIIc. Colonies up to about 500 $\mu$  deep, blue green  
 producing abundant mealy masses of conidia  
 (not crusts or solid adherent masses); conidia mostly globose about 3 $\mu$ , occasionally  
 larger.....*P. griseo-roseum* Di-  
 ereckx, 142.
- XLIIId. Colonies rather thin close textured; conidia  
 2.6 to 3.2 $\mu$ .....*P. notatum* Westling,  
 144.
- XLIIIe. By description placed near to *P. notatum*.....*P. virescens* Bainier,  
 145.
- XLIII f. Colonies up to 1000 $\mu$  deep; becoming slowly  
 red below; conidia up to 3 or even 3.5 $\mu$ .....*P. citreo-roseum* Di-  
 ereckx, 146.
- XLIIIg. Colonies.....*P. meleagrinum* Bi-  
 ourge, 147.
- XLIV. Colonies shallow (100 to 300 $\mu$ ) near the margin  
 become 500 to 1500 $\mu$  in center; reverse and agar yellow to reddish shades;  
 drops yellow; conidia 3.5 to 4 $\mu$ .....*P. cyaneo-fulvum* Bi-  
 ourge, 148.
- XLIV. Colonies up to 1000 $\mu$  deep, bluish gray green,  
 to sage green, to brown; drops colorless or pale yellow;  
 conidia 3.5 to 5 by 2.5 to 4 $\mu$ .....*P. brunneo-rubrum*  
 Dierckx, 149.
- XLIV. Colonies about 400 to 600 $\mu$  deep; conidia usually showing fairly general ellipticity with  
 long axis 4 to 5 $\mu$ .....*P. baculatum* Westling, 150.
- L. Velvety sections with more or less erect conidiophores with walls mostly rough or pit-

ted, and penicilli usually showing sterigmata, metulae and branches of one or more series in a compact brush or broom.....LI.

LI. Colonies narrowly growing.....LII.

LI. Colonies spreading broadly.....LV.

Restricta (*P. puberulum* and allies)

- LIIa. Colonies wrinkled and buckled thin felts; bluish glaucous; yellow to brown below; conidia 3 by 2 to 2.5, later 3 to 3.5 $\mu$  subglobose.....*P. casei* Staub, 155.
- LIIb. Colonies velvety blue green to sordid green; suggested as synonymous.....*P. biourgei* Dierckx, 156.
- LIIC. Conidiophores 100 to 200 $\mu$  long; conidia about 4 to 4.2 $\mu$ .....*P. puberulum* Bainier, 157.
- LIId. Colonies bluish gray to gray, conidia 3 to 3.5 $\mu$  rough.....*P. melinii* Thom, 158.

LV. Colonies broadly spreading with broad margin cobwebby (arachnoid).....Stellata—*P. roqueforti* series LVI.

LV. Colonies velvety but lacking the arachnoid margin.....LX.

Stellata or *P. roqueforti* Series

- LVIa. Colonies with broadly arachnoid margin in bluish green to green shades, with reverse mostly showing yellow to greenish colors; conidia 4 to 4.5 to 5 $\mu$  (occasional larger ones).....*P. roqueforti* Thom, 160.
- LVIb. Conidia 4 to 6 $\mu$ .....*P. stillon* Biourge, 161.
- LVIc. Conidia 3 to 4 $\mu$ .....*P. atro-viride* Dierckx, 162.
- LVID. Cheese mold vaguely described. Probably synonymous with *P. roqueforti* Sopp...*P. aromaticum* I Sopp, 163.
- LVIe. Conidia extra large 5.5 by 4.5 $\mu$  or 7.2 by 6.5 $\mu$ ...*P. roqueforti* var *megalospora* n.n. Dierckx, 164.
- LVIIf. Reverse of colonies green.....*P. roqueforti* var. *Weidemannii*, Westling, 165.
- LVIg. Conidia globose 3 to 5 $\mu$ .....*P. Biourgei* Arnaud, 168.

- LVIh. Distinguished by lack of rosy color below;  
colors potato brown.....*P. Weidemanni* var.  
*fuscum*, Arnaud, 169.
- LVIIi. Conidia 6 to 8μ.....*P. virescens* Sopp,  
170.
- LVIj. Colonies spreading quickly pale gray green;  
conidiophores very short with coarsely gran-  
ular walls; conidia about 4μ.....*P. suavolens* Biourge,  
171.
- LVIk. Colonies blue green; conidia up to 6.5 by 5.5  
to 5.8μ.....*P. gorgonzola* in Bi-  
ourge, 167.
- LVIIn. Colonies blue-black, to black brown; conidia  
4 to 6μ.....*P. atroviridum* Sopp,  
173.
- LVIIm. Colonies bluish-green; conidia 5μ.....*P. roqueforti* Sopp,  
var. a, 174.
- LVINn. Colonies bluish green; conidia 6μ.....*P. roqueforti* Sopp  
var. b, 174.
- LVIo. Colonies bluish green to yellowish green gray-  
ish finally brown; conidia 4 to 7μ.....*P. aromaticum* Gam-  
melost Sopp, 175.
- LVIp. Colonies bluish gray to gray brown; conidia  
yellow brown 7 to 8μ.....*P. griseo-bruneum*  
Sopp, 176.
- LVIq. Colonies pale green to dark green; vesicular  
cells abundant; conidia 3.7μ.....*P. vesiculosum* Bai-  
nier, 177.
- LX. Colonies broadly spreading, lacking the arach-  
noid margin of the *P. roqueforti* series (cf.  
Radiata—with smooth walls) walls rough....LXI.
- LXI. Velvety, spreading, with asperulate walls,  
azonate or hemizonate.....LXII.
- LXI. Zonate.....LXV.
- LXII. Colonies velvety, spreading, with walls rough,  
fairly wide margin; conidia 4.2μ.....*P. asperulum* Bai-  
nier, 180.
- LXV. Colonies deeply velvety and ridged zonate in  
age but showing the rough appearance of  
fasciculation when young hence placed in  
Fasciculata-aeruginosa.....*P. flavo-glaucum* Bi-  
ourge, 238.

*Sub-section 1. Elliptica-Magna.* An aggregation of species with large definitely and persistently elliptical conidia and generally velvety colonies. Biourge's *Fragilia* mostly belong here.

110. *P. niveum* Sopp. Monogr., pp. 182-184, Taf. XXIII, fig. 16. 1912.

Colonies on meat-peptone-sugar-gelatine white, Mucor-like, with fibrous, weakly growing, loose mycelial network, chalk-white, with white mealy conidial areas, fairly thick in age; hyphae coarse; reverse of colony uncolored but substratum colored purple brown; gelatine slowly liquefied; odor none; conidiophores coarse, with true penicillate branching; sterigmata long, straight, tapering to long slender tubes; conidia abundant, oval, 9 by 18 to 20 $\mu$ , with chains breaking up easily, so that rarely more than 2 or 3 remain attached; perithecia not found.

Species found as a parasite on *Polyporus zonatus* and apparently more closely related to *Acaulium* than the typical *Penicillia*. Sopp regards *P. olivaceum* (= *P. digitatum* Sacc.) and this form as doubtfully included in the genus.

Cultures grew best at 20°C., with minimum at 5° and maximum at 35°C., and grew well in milk, upon potato, rice and bread. Conidia remained viable more than three years.

No species with this size of conidia has been seen by us. It has been included here because Sopp compared it to *P. digitatum* ("*olivaceum*") although he also compared it to *Acaulium* (*Scopulariopsis*).

111. *P. digitatum* Sacc. In *Mycotheca Italica*, No. 986, Herbarium U. S. Department of Agriculture; in *Sylloge Fungorum*, Vol. IV. p. 79; in *Fungi Italici*, No. 894.

Possible Synonym:

*P. olivaceum* Wehmer. Beitr. z. Kennt. einheim Pilze, pp. 73, t. II, Jena, 1895.

? *Mucor caespitosus* L. In *Species Plantarum* (1753), II, p. 1186 based upon Micheli tab. 91, fig. 3.

? *Monilia digitata* Fries. *Systema Mycologicum*, III, p. 411. 1829.

Micheli, *Genera Plantarum* (1729), pl. 91, fig. 3.

Linnaeus, *Species Plantarum* (1753), II, p. 1656.

Persoon, *Observationes*, p. 41.

Persoon, *Synopsis Fungorum*, p. 693. *Monilia digitata*.

Compare:

Biourge, *La Cellule* 33: fasc. 1, pp. 210-212; Col. Pl. V, Cart. 30; Pl. VIII, fig. 46. 1923.

Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bull. 118: pp. 31-33, fig. 3. 1910.

Description as given by Thom (fig. 29, 30).

Colonies on sugar gelatin and potato or bean agar grayish olive, irregularly shaped from the unequal growth and branching of rather few hyphae, aerial portion consisting only of very short conidiophores and conidia; reverse of colony commonly shows brown to black colors; conidiophores rising directly from the substratum, 30 to 100 by 4 to 5 $\mu$ ,

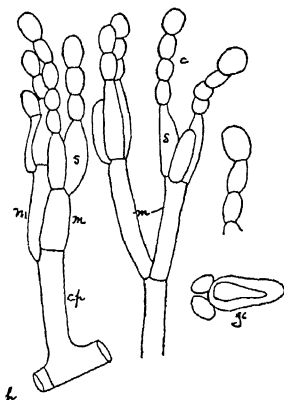


FIG. 29. *P. digitatum* Sacc.: *cp*, short conidiophore; *m*, branches or metulae; *s*, sterigmata; *c*, conidia; *gc*, germinating conidium.



FIG. 30. *P. digitatum*: Diagrammatic radial section of young colonies on Czapek's solution again showing the very thin layer of spreading penicilli.

usually very short; penicilli a few tangled conidial chains up to 160 $\mu$  in length, borne upon sterigmata 13 to 16 by 3 to 4 $\mu$ ; conidia cylindrical to almost globose, 4 to 7 by 6 to 8 $\mu$  (at times 6 by 10 $\mu$ ), often uneven in size and shape in the same chain; colonies do not liquefy sugar gelatin except at times, partially, in cultures three weeks old or more. Litmus reaction acid. Grows readily on organic media, but shows a very pronounced affinity for media with high percentages of sugar, in which it produces a strong odor.

Colonies on Czapek's solution agar, and wort agar, in olive shades



varying toward green but not truly green, and producing an aromatic odor. The following measurements are given by Biourge: conidiophore about  $5\mu$  in diameter, usually very short; penicillus various about  $60\mu$  long with all walls smooth; branches in two's or three's, appressed or divergent  $15$  to  $50\mu$  by  $3$  to  $4.5\mu$ ; typical metulae not found; sterigmata  $11$  to  $28$  by  $3.5$  to  $5\mu$ , in two's, three's, or four's; conidia varying greatly in size and appearance  $3$  to  $5$  by  $2.5$  to  $3\mu$  at first, then  $6$  to  $8$  by  $4$  to  $6$ , again almost globose  $10$  to  $11\mu$ , or elliptical up to  $12$  to  $21$  by  $6$  to  $9\mu$ .

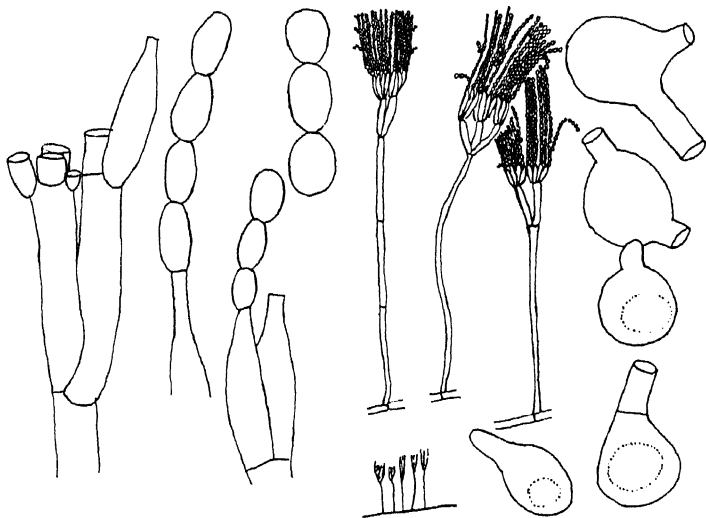


FIG. 31. *P. oxalicum* Currie and Thom: Showing contrasting habit to *P. digitatum* with closely similar morphology.

Saccardo's specimen in *F. italici* no. 894 was seen and is near enough to type material to be positive in identifying his species.

Cosmopolitan upon citrus fruits, distinguished from *P. italicum* by the sharp contrast of its olive color with the blue of the other. Collected in Hanover and verified by Dr. C. Wehmer. Received from Prof. P. H. Rolfs in Florida. Seen upon decaying oranges everywhere. Pure cultures can always be secured from the common market fruits.

The olive rot of oranges is constantly encountered in handling citrus fruits. It spreads rapidly over the surface of the orange with the production of a dense powdery layer of the typical olive colored conidia.

Such oranges if exposed to the air dry up rapidly, shrinking in size to become eventually hollow shells in contrast to the effect of *P. italicum* which produces a soft rot from which the orange quickly loses its shape, flattening into a slimy mass.

In our culture experience *P. digitatum* has been found only in connection with citrus fruits but Thakur and Norris working in India report it also from soil.

111a. *P. digitatoides* Peyronel in I germi atmosferici dei funghi con micelio, page 22, Padova. 1913.

Peyronel admits that this species was only a cultural form of *P. digitatum*

112. *P. digitatum* Sacc. var. **Californicum** n. var.

Variety differs from the species only by entire absence of green color: colonies white.

Type Thom and Church no. 4747 contributed by Dr. H. S. Fawcett from Riverside, California. He reports the variety equally destructive in attacking oranges as the usual olive green form.

113. *P. olivaceum* Wehmer. Beitr. z. Keuntn. Einh., Pilze II, 1, pp. 73-76; Taf. I, fig. 2, Taf. II, fig. 11-15; Jena. 1895.

Synonym: *P. digitatum* Saccardo, which see.

Wehmer's description and figures of this species are good and his name for it is so apt and descriptive that it is unfortunate that it must be replaced by that given by Saccardo which was considered and refused by Wehmer for members of this group (loc. cit., p. 69). Our conception of the species was obtained from cultures made directly from rotting oranges in Wehmer's laboratory and compared to specimens actually distributed by Saccardo, thus leaving no room for doubt as to identity.

114. *P. olivaceum* var. *italicum* Sopp. Monogr., p. 179, Taf. XIX, fig. 133; Taf. XXIII, fig. 29. 1912.

Synonyms: *P. olivaceum* Wehmer; *P. digitatum* Sacc. q.v.

Sopp gives only the following: Colonies spreading broadly in the substratum with a radiating fibrous appearance and slowly producing scattered short conidiophores and conidial masses characteristic of *P. digitatum* Sacc.; Conidia 5 to 6 by 9 to 15 $\mu$ : these measurements probably covered conidia which were actually in the early stages of germination.

115. *P. lanoso-grisellum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 196–198; Col. Pl. III Cart. 310; Pl. V, fig. 25. 1923.

Colonies on wort gelatine deeply lanose, covering the whole surface of the medium, gray green with a wide deep white marginal zone; coremia none; reverse pallid ochraceous; odor none; gelatine not liquefied; penicillus figured as a main axis with a terminal verticil of either metulae or sterigmata with perhaps one short appressed branch and all about  $35\mu$  long, or a similar terminal mass and 1, 2 or 3 secondary masses from upper nodes of the stalk making a total of 90 to  $100\mu$  in length; branches irregular; metulae 14 to 25 by 3 to  $5\mu$ , in pairs; sterigmata 10 to 18 by 3 to  $5\mu$ , in threes or fours deciduous; conidia 5 to 8 by 3.5 to  $4.2\mu$  to subglobose  $5.5$  by  $5\mu$ , 1-guttulate.

Biourge's no. 310 (not seen by us) is described as having sterigmata as in *P. camemberti* and conidia long cylindrical as in *P. italicum*, with all parts of the penicillus falling away easily and conidia that die quickly. His culture was early lost. It was obtained in the Mediterranean region and cultured by Dierckx.

While not seen by us, the description suggests a possible variety of *P. digitatum* or a form with nearly related morphology.

116. *P. olivaceum* var. *discoideum* Marchal.

Marchal described under this name an organism on prunes which produced olive green discoid masses which suggest *Metarrhizium* (see no. 353, Chap. 19).

117. *P. olivaceum* (?) Sopp. Monogr. pp. 176–179, Taf. XIX, fig. 133, 135, and Taf. XXIII, fig. 29, 30. 1912.

Without citing Wehmer as the describer of *P. olivaceum*, Sopp attributes the name used to the label of a culture from Kral with which his own isolations were compared.

118. *P. olivaceum* var. *norvegicum* Sopp. Monogr., p. 177–179, Taf. XIX, fig. 135; Taf. XXIII, fig. 30; cited by Biourge Monogr. p. 211 as var. "*norvégienne*," and apparently intended to be the basis of *P. wurceburgense*. In table p. 136 as "*var norvegica*."

Colonies on meat-peptone-sugar-gelatine clear green to olive green with fibrous appearance, almost submerged mycelium, spreading over the whole substratum; reverse at first white then greenish; hyphae coarse; odor moldy (?); upon rice a perfume (ethereal or alcoholic?); conidio-phores scantily produced, coarse, short, septate, figured as few branched

and producing verticils of few long sterigmata tapering to long tubes and cutting of oidium-like conidia; conidia 6 to  $10\mu$  by 12 to  $18\mu$  (see table, page 137) on p. 178 the indication for "by" omitted and the inference is probably correct that 12 to 18 are maximum long axes observed probably in germinating spores; perithecia not found.

Species found upon cellar earth. Cultures grew best at  $20^{\circ}$ , with minimum at  $5^{\circ}$  and maximum at  $33^{\circ}\text{C}$ . Although colonies developed rapidly upon broth and potato, this species was in general not readily adapted to laboratory media.

Conidia remained viable more than three years.

We have not found a form with the measurements given by Sopp in this species.

119. *P. wurceburgense* is proposed by Biourge, Monogr. p. 211 and figured in Pl. VIII, fig. 47 and colored plate V, No. 156, for a culture distributed by Kral in 1913 as coming from Würzburg and having the conidial measurements given by Sopp for his *P. olivaceum* var. *norvegicum*.

While the description of Sopp, and Biourge's notes leave the identification doubtful, the cultures referred to by them probably indicate a species found in European soil similar to *P. oxalicum* Currie and Thom but with larger conidia. The species may eventually be rediscovered and described.

120. *P. oxalicum* Currie and Thom. Jour. Biol. Chem. 22, no. 2, p. 289, fig. 1. 1915. See fig. 31.

A soil inhabiting species producing oxalic acid rapidly in sugar media. Colonies ivy-green velvety, broadly spreading, with reverse pale yellow, and agar uncolored or only slightly colored; conidiophores up to  $200\mu$  by  $3.3$  to  $5.4\mu$ , producing a verticil of 2 to 3 metulae, verticils of closely parallel sterigmata 10 to 14 by  $2.5$  to  $3.5\mu$ ; conidia cylindrical then elliptical, uneven in size from  $3$  by  $2\mu$  when first formed to a maximum of  $5$  by  $3.5$  when ripe and becoming subglobose  $7$  to  $10\mu$  in diameter in germination, in long chains packed in close columns. In favorable media producing great masses of conidia which break off readily.

Cultures: Type no. 103 from soil, Thom; also contributed from Manchuria and from many places in America, as indicated in the following selected list; 4322.261.1 Hoffer, Illinois and Indiana corn; 4315 A; H.4312.71518.1. Wood—Madison; 4295 Shapovalov; 4279—H Saito—Manchuria; 4920.10 from Arlington farm, Virginia.

and Perrier used the name for a *Citromyces* which was so inadequately described that no attempt at identification is possible hence the name may be allowed to lapse with the generic name *Citromyces*.

A variation of this species (no. 4913) received from Helen Johann at Madison, Wisconsin, is discussed by her as injuring corn seedlings (*Phytopathology* 18: 239-242. 1928). Notes on this strain are added as showing a range of cultural characters in the species:

Colonies on Czapek's solution agar, broadly spreading, azonate, or showing a trace of zonation when viewed from below, velvety, in color a dark dull green shade near sage green to slate olive (Ridgway XLII.) but with a slightly bluish tinge near the margin; reverse colorless or slightly yellowed: odor, none; drops not seen; conidiophore about 100 to 200 by  $3\mu$ , smooth; penicillus either monoverticillate or mostly with one branch or metula (sometimes two), usually appressed and with the conidial chains forming one or two columns often  $500\mu$  or longer and commonly 10 to  $15\mu$  in diameter, forming tangled masses over the whole colony which break off when struck or tapped; metulae about 20 by  $3\mu$ , in groups of 2 to 3; sterigmata up to 12 to  $14\mu$  by about  $3\mu$ , taper pointed; conidia elliptical about 5 to 6 by 3 to  $4\mu$  mostly 5 to 5.5 by 3 to  $3.5\mu$ .

Numerous cultures of this species have been seen upon the plates made from soil in the Soil Microbiology laboratories at Washington.

121. *P. duponti* Griffon and Maublanc. (See Griffon, R., et Maublanc, A. Deux Moisissures thermophiles) *Bul. Soc. Mycol. France* 27: 68-74, fig. 4-8. 1911.

Colonies on bean decoction with agar and upon potato, white then slowly pale gray green, finally dark brown, with aerial creeping hyphae hyaline, very fine (2 to  $3\mu$  in diameter), septate, bearing conidiophores at irregular intervals as short branches; conidiophores 10 to  $30\mu$  in length, ascending, unseptate or 1-2 septate, with apex branched (as figured) variously to produce branches or metulae, terminating in small verticils (figured as 1 to 3 or 4) of sterigmata about  $10\mu$  in length, bearing conidial chains; conidia globose-ovoid, subhyaline, smooth, mostly 2 to 5 by 1.5 to  $4\mu$ , occasionally up to  $9\mu$  in long axis; chlamydospores develop here and there in the submerged hyphae as enlarged thick walled cells.

Species obtained from fresh manure which heated naturally and from wet hay in the autoclave at  $50^{\circ}$ . Cultures grew between 25 and  $60^{\circ}\text{C}$ . with an optimum at  $45^{\circ}$  to  $50^{\circ}\text{C}$ .

*Sub-section 2. Velutina-divaricata*: Colonies velvety or velutinous but with each branch in the penicillus diverging like a monoverticillate penicillus (fig. 32).

In addition to the species and strains included here certain species in Zaleski's series which are apparently velvety or so nearly so as to be sought here have been placed among the soil organisms in the Section Lanata-Divaricata. See *P. Janczewski* etc.

\* *Conidia definitely elliptical (less than  $5\mu$  in long axis).*

125. *P. rubens* Biourge. Monogr. La Cellule 33: fasc. 1, p. 265; Col. Pl. XI, Cart. 407; Pl. XIX, fig. 111. 1923. See fig. 33.

*P. rufescens* incorrectly on Col. Pl. XI. Carton 407.

Colonies on wort gelatine, wrinkled, obscurely bluish glaucous, then grayish brown; coremia none; reverse orange to fulvous; wort gelatine not liquefied; (?) odor none; conidiophore 2.8 to  $3\mu$  in diameter, here and



FIG. 32. The divaricate type of *Penicillus*: An extreme example



FIG. 33. The *P. rubens* type of colony: Diagrammatic radial section (magnified 25 times) showing the thickening of the mycelial mass toward the center and under the yellow drops.

there dilated to  $5\mu$ ; penicillus about  $25\mu$  long, with all walls smooth; figured as either monoverticillate or biverticillate with divergent metulae; metulae 10 to 16 by 2.2 to 2.5, commonly in threes, sometimes 1 or 2 only; sterigmata 10 to 14 by 2.5 to  $3\mu$ , in verticils of 2 to many; conidia oblong 3.5 to 5 by 2.8 to  $4\mu$ .

Biourge's type culture no. 407 (our no. 4733.110) when grown on Czapek's solution agar produced colonies velvety not over  $200\mu$  deep, radiately wrinkled almost crenulate at edge and umbonate in center; mycelium a tough dense felt; blue green becoming brown in age; reverse yellow at outer margin to reddish orange in center; agar more or less yellowed; conidiophores short, smooth; penicillus consisting of main axis, terminal verticil of divergent metulae with often a secondary fruit-

ing branch lower down and verticils of sterigmata producing columns or very closely parallel chains of conidia; conidia 3.5 to 4 by 2.8 to 3.3 $\mu$ , with some persistent ellipticity.

Several cultures having the characters given by Biourge for *P. rubens* have been received including no. 5016.10a from a decaying *Strobilomyces*; 4975.119 from decaying sugar beets, Utah.

126. *P. rubrum* (?) Grassberger-Stoll. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 172-174; Col. Pl. IV, Cart. 164; Pl. VI, fig. 33. 1923.

Not *P. rubrum* Stoll.

Colonies on wort gelatine, with a thin aerial felt, almost velvety, at first blue green, then greenish brown to brown, occasionally and sporadically producing red nodules; reverse citrine, then yellow, conidiophore 3 to 4 $\mu$  in diameter, with all walls smooth; penicillus 25 to 30 $\mu$ , figured as a central axis and terminal verticil of metulae alone, or with branches from the nearest node, or nodes bearing secondary groups of metulae; metulae 9 to 12 by 2 to 2.5 in verticils of 2 to 4; sterigmata 8.5 to 10 by 2 to 3 $\mu$ , in verticils of 2 to 7; conidia elliptical 3 to 4 or even to 5 by 2.8 to 3.5 $\mu$ .

Biourge's culture no. 164 (our 4733.111) is attributed to Kral and not reported as found in nature. While very slight differences may be seen in parallel culture, this organism is not more than a variety or strain of *P. rubens* Biourge. It is not *P. rubrum* Stoll. To present this material as fully as possible our notes on Biourge's culture follow: Colonies in Czapek's solution agar, velvety forming a layer 150 to 250 $\mu$  deep, radiate wrinkled, with central areas raised, rather bright green near "stone" green (Ridgway XLII) and again bluish gray green on the same plate, with margin white consisting of a narrow submerged area merging gradually into a velvety green area; reverse citrine, drops numerous pale citrine; odor, none or indefinite; conidiophores ascending rather than erect up to 100 or even to 200 $\mu$  by 2 to 2.5 (even to 3 $\mu$ ), with walls smooth; penicillus various consisting of a single terminal group of branchlets unequal or equal and metula-like, usually diverging, occasionally with sterigmata in the same verticil, and commonly other branches of varying complexity at the second or third septum, various in length and in aggregation of elements; branches or metulae commonly unequal; sterigmata commonly 7 $\mu$ , occasionally larger and up to 10 $\mu$  by 2 to 2.5 $\mu$ ; conidia 3.5 to 4 by 2 to 3 $\mu$ , with persistent ellipticity, in chains parallel or somewhat diverging (not in columns), more or less tangled in age.

127. *P. obscurum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 267-269; Col. Pl. VIII, Cart. 120; Pl. XIV, fig. 80. 1923.

Colonies on wort gelatine, spreading, at first blue green (coeruleus) then dark green, becoming brown, fuscous or fumose; coremia none; reverse sordid or more or less yellowish brown; odor none; conidiophore 2 to  $3.5\mu$  in diameter; penicillus from 10 to  $15\mu$  long when monoverticillate, to 25 to  $30\mu$  in the biverticillate form in which the branches diverge, described as with walls smooth (but in the figures all walls including the sterigmata are marked as if asperulate); metulae 16 to 20 by 2 to  $2.5\mu$  from one to three in number; sterigmata 8 to 11 by 3 to  $3.5\mu$ , in verticils of 4 to many; conidia elliptical 3 to 4 by 2 to  $3.4\mu$  to globose 4 to  $5.6\mu$  with the younger 5 to 10 conidia in the chain smooth, the outer or older ones rough or asperulate.

Our no. 4733.91 received from Biourge as his type no. 120, does not show rough conidia of the size described nor markings on the walls as indicated in Biourge's figures. This organism can be placed close to *P. corylophilum* as indicated in Biourge's Monograph. Only a minority of the penicilli in this culture show metulae. The monoverticillate type predominates in number although the branching form may be frequent enough to justify putting it in the Asymmetrica. One is compelled to believe there is some mistake either in description or in placing of the species. It is placed here because of the figures given but disregarding the culture.

128. *P. atramentosum* Thom. Emended from U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 65, 66; fig. 24. 1910.

Colonies upon gelatin or upon potato or bean agar velvety, narrowly growing, azonate bright green, with aerial part mostly of simple conidiophores mixed in older parts with branching aerial hyphae; reverse more or less yellow; conidiophores 240 to  $400\mu$  usually about  $300\mu$  long; penicilli either a single one-sided verticil of metulae or main axis and one or two branches producing similar verticils of metulae producing diverging columns of conidia 100 to  $200\mu$  long; sterigmata 8 to  $10\mu$  long, closely packed in the verticil; conidia 3.5 to 4 by 2.5 to  $3\mu$  occasionally larger up to  $4.5\mu$  in long axis and sometimes subglobose, homogeneously green or yellowish green in color, in germination swelling to 6 or  $7\mu$  in diameter and producing single tubes. Colonies liquefy sugar gelatin, producing alkaline reactions, digest milk and color potato agar containing sugar an inky black whence the name.

The type Thom no. 38 was found as a contaminant upon a Camembert



cheese imported from France. Transfers of it were sent to several European correspondents and our type culture was lost. Biourge, however, included the species in his monograph and returned his transfer of our culture which appears to be true to description.

Biourge (in Monogr. La Cellule 33: fasc. 1, p. 260-262; Col. Pl. IX, Cart. 161; Pl. XIV, fig. 84, 1923) describes it as follows: Colonies on wort gelatine mostly velvety, dull bluish green, then dark green, dark olive, at length olive brown; coremia none; reverse yellow with sordid hyacinth tones, through browns to almost black in age; odor peculiar suggesting amyl acetate; gelatine liquefied; conidiophore 2 to  $4\mu$  in diameter, very irregular in length, with all walls smooth; penicillus from a simple verticil of metulae about  $25\mu$  long, with the main axis occasionally prolonged to form a second superposed verticil hence 35 to 55 in total length; branches rare; metulae 8 to 12 with apex enlarged; sterigmata 8 to 12.8 by 3 to  $3.8\mu$ , in verticils of 2 to 6; conidia 3.5 to  $4.8$  by  $2.5$  to  $3.5\mu$ .

Biourge no. 161 (our no. 4733.3) is noted by Biourge as conforming to Dierckx's "Diversiramosa," in which the verticil of metulae is asymmetrical—1-sided. Grown upon Czapek's solution agar, 4733.3 produced colonies azonate dull dark green, with marginal 1 to 2 mm. white, and a trace of bluish in the newer conidial areas, with reverse in purple brown, sterigmata deciduous in mounts, and conidia about 4 by  $3\mu$ .

*Conidia globose or subglobose. Chains of conidia divergent.*

131. *P. chloro-leucon* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 270-271; Col. Pl. VIII, Cart. 45; Pl. XIV, fig. 79. 1923.

Colonies on wort gelatine, velvety more or less wrinkled, at first bluish green then dark bluish green, to dark gray, finally brown, coremia none; reverse at first very pale green, then sordid yellowish brown; odor none; conidiophore 2 to  $4\mu$  in diameter; penicillus 15 to  $35\mu$  long, with all walls smooth, figured as a verticil of sterigmata or a terminal group of 2 to 4 branches or metulae more or less diverging; metulae 15 to  $20\mu$  long, two, three, four or none; sterigmata 9.5 to 13 by  $3\mu$ , in groups of 2 to 8  $\mu$  conidia ellipsoid to globose, 3 to  $4.5$  by  $2.7$  to  $3$ .

Biourge's type no. 45 (our no. 4733.30) grown upon Czapek's solution agar apparently velvety but with some creeping hyphae, about  $200\mu$  deep, dull deep green to gray green spreading broadly, with white margin 1 to 1.5 mm. wide; reverse uncolored or slightly rose purplish in deeper areas, in age more or less brownish; conidiophores 2 to  $2.5\mu$  in diameter,

very short branches of creeping aerial hyphae, or terminal on such hyphae rarely more than  $100\mu$  long, with cells commonly enlarging from base to apex, penicillus a single verticil of sterigmata, or a central axis and 1 to 3 branches or metulae from the topmost node, unequal 10 to  $20\mu$  long in the same group, each with swollen apex bearing a verticil of a few sterigmata 7 to 10 by 2 to  $2.5\mu$ , bearing long parallel chains of conidia which become tangled in age; conidia subglobose about  $4\mu$ .

Two other cultures 4742C, from England, and 4207.5 from cranberries produced in the Rocky Mountain States appear to belong here.

*P. Westlingi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 473, 474; Taf. 54; Zaleski no. 253b.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, slow growing becoming 24 to 26 mm. in diameter in twelve days, with liquefaction of the gelatine beginning about the twelfth day and proceeding rapidly, commonly velvety, azonate, with distant radiate wrinkles in the outer part of the colony and central area more or less elevated sometimes conspicuously; margin about 1 mm. wide, in the older but still growing colony; in color blue green C.d.C. 367, 422, 423, to green 343, becoming dark shades of orange brown such as 140, 114, 115 in age; reverse and liquefied gelatine pale yellow shades such as 203A 203B, 171, 162, 157, 152; drops small, uncolored, very numerous in the growing conidial areas; odor none; conidiophores 200, 300 to 500, or even  $700\mu$  long by  $2.5$  to  $3.5\mu$ , species enlarged capitate, all walls smooth, sparingly branched; penicilli 20 to  $25\mu$ , or when branched 40 to  $65\mu$  long, with sterigmata and metulae often mixed in the verticil; branches rare, 10 to  $40\mu$  long and only one or two at a node; metulae varying in length and asymmetrically arranged in this verticil 8 to 16 by  $2.5$  to  $3.5\mu$ , with apices enlarged to  $4.5\mu$ , about 3 to 6 in the group; sterigmata about 9 to 10 by  $2.3$  to  $2.8\mu$ , in verticils of about 6 to 8; conidia about  $2.3$  to  $2.8\mu$ , smooth, subglobose to globose, with connective evident.

Habitat: Species isolated from earth in the pine forest "Długa Goslina" near Posnan in Poland.

From the microscopic structure, Zaleski notes that Biourge placed this species in the series with *P. chrysogenum* in the Radiata. No such placing is possible from the description given, however (T) Zaleski himself puts it in "*Biverticillium Dierckx* subsec. 3. Radiate undulata." Our notes follow: Colonies upon Czapek's solution agar spreading fairly widely, in the outer areas plane, velvety or nearly so, becoming floccose and deeply wrinkled or convoluted in the central area; conidial areas

bluish gray green, gray green becoming dull olive green in age; reverse colorless or more or less yellow; drops, yellow, in central area, seen in cultures at 20°C., not at 30°C.; conidiophores 100 to 200 $\mu$  near the margin as trailing hyphae up to 500 or longer in central area; penicilli consisting of a terminal group of diverging more or less unequal branchlets (or metulae?) with or without a secondary group from the next septum and monoverticillate branchlets scattered along the main axis; sterigmata 7 to 8 $\mu$  long; conidia 2 to 2.5 $\mu$  (to 2.8 $\mu$  according to Zaleski); in parallel to divergent or tangled chains.

Culture no. 5010.28 received as type from Baarn in July, 1928, appears to be correct. This organism in culture often suggests *C. subtilis* of Bainier and Sartory which is placed in Chapter XIII on account of its branching conidiophore with each branch bearing a monoverticillate penicillus.

*Chains of conidia in columns and the columns divergent.*

133. *P. corylophilum* Dierckx. Soc. Scientifique Bruxelles 25: p. 86. 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 266-267; Col. Pl. IX, Cart. 78; Pl. XIV, fig. 83. 1923.

Colonies on wort gelatine spreading, with narrow white margin, blue green becoming dark in age almost black, or fuscous; coremia none; reverse yellow green to orange shades; odor none; conidiophore about 50 by 2 to 3.5 $\mu$ , figured as arising from creeping hyphae; penicillus 20 to 45 $\mu$ , with all walls smooth, figured as monoverticillate or asymmetrically biverticillate; metulae 10 to 23 by 2.5 to 3.5 $\mu$ , in pairs or asymmetrically in threes; sterigmata 9.5 to 13 by 3 to 3.5, in verticils of 2 to 6; conidia 3 to 3.6 (5 to 6) (?CT) by 2.5 to 3.5 (4.5) $\mu$ .

Biourge no. 78 (our no. 4733.42), presumably type, grown upon Czapek's solution agar spreading broadly, thin in mass, velvety, 100 to 200 $\mu$  deep with more or less trailing and creeping hyphae and with a deep white overgrowth in center, with margin white, uneven, with radiating lines of conidiophores leading out into the sterile area; dull green in the conidial area; reverse colorless to greenish to bluish black; conidiophores short, penicillus citromyces-like or a verticil of metulae each producing a columnar mass of conidia; sterigmata about 9 to 10 $\mu$  long, with fairly sharp point; conidia mostly globose 2 to 3 $\mu$ , some 3.5 to 4 $\mu$ , considerable irregularity in size.

Several other forms apparently belonging here have been studied. These include no. 4658.118.4, 1.3 and 38.8 from Putterill at Cape Town,

4053.18 from New Jersey soil, 4833P from honey bees as studied by Burnside, 4777.39 from Pribram. Biourge's No. 120 (4733.91) as received labeled *P. obscurum* is near *P. corylophilum*.

134. *P. umbonatum* Sopp. Monogr., pp. 196-198; Taf. XXI, fig. 148; Taf. XXIII, fig. 40. 1912.

Colonies on meat-peptone-sugar-gelatine, gray-green, with a marked projection or umbo in center due to buckling of the mycelium, and producing small disk-like colonies showing in figure 40, as with broad sterile marginal zones; reverse colorless or with a tinge of Naples yellow, especially in center, but colors potato black; hyphae comparatively coarse; odor strong, moldy, peculiar; conidiophores small, slender, with branches producing either monoverticillate conidial apparatus or verticils of short or long metulae from a vesicular apex, each producing a monoverticillate conidial mass; conidia 3 to 3.5 $\mu$ ; perithecia not found.

Species found upon various mushrooms, especially *Clitocybe nebularis*, in Norway. Cultures grew best at 20° with minimum at 1° and maximum at 35°C., but in general grew weakly upon Sopp's media, but grew fairly well upon urine, on rice, on tannic acid solution, and oak chips. Conidia remained viable more than three years. No one since Sopp has surely identified this species.

135. *P. steckii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 469, 470, 471; Taf. 50; Zaleski no. 1631b.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 28 to 30 mm. in diameter in twelve days, quickly and completely liquefying the gelatine; surface uneven, velvety or slightly floccose, azonate, thrown into a few radiate wrinkles with a raised central area; margin white, velvety, 2 to 3 mm. or even 4 mm. wide when young; in color blue green 367 to green 348, to orange brown shades such as 169 in age; reverse in pale yellow shades 171, 166, 161; drops few uncolored, in the center or scattered over the whole surface; odor none; conidiophores varying 100, 200, 300 or even 400 $\mu$  long, by 2.2 to 3 $\mu$ , with walls smooth, simple or sparingly branched; penicilli about 18 to 24 $\mu$  long, as figured monoverticillate or divaricately and asymmetrically biverticillate; metulae when present about 10 to 14 or even 18 $\mu$  by 2.5 to 3.5 $\mu$ , usually in unsymmetrical verticils of 4 to 6, shown in figures as divaricate, and with vesicle-like apices; sterigmata about 9 to 10 by 2.3 to 2.8 $\mu$ , with a definite more or less coarse tube in verticils of 5 to 10, or occurring singly on the main axis or in the same verticil with

metulae; conidia 2.2 to 2.5 $\mu$  (even to 2.8 $\mu$ ), smooth, subglobose to globose; Habitat: Species isolated from earth in pine woods in square "369" of the forest "Puszcza Bielowiecka" in Poland.

Zaleski reports that Biourge placed this in the "diversiramosa" of Dierckx with *P. atramentosum* Thom while Zaleski himself puts it in "Biverticillium Dierckx subsec. 3. Radiate-undulata." Our notes follow: Type strain growing better about 20° than about 30°C., and much better upon wort than upon Czapek; colonies upon Czapek's solution agar, becoming 18 to 30 mm. in diameter in ten days, velvety increasing from very shallow at the margin to 500 $\mu$  in the central area, broadly but indistinctly zonate, with marginal 1 to 2 mm. white passing through pale bluish to a central area grayish blue green (Ridgway XLVIII); reverse uncolored at first more or less zoned with faintly yellowish bands in age; odor, none; conidiophores up to 500 $\mu$  by 2 to 2.5 $\mu$  with apex inflated and with walls smooth; penicillus sometimes a single verticil of sterigmata, mostly a verticil of more or less unequal branches (or metulae?) each bearing a verticil of sterigmata producing a long columnar mass of conidial chains, with the columns of conidia mostly separate to the base; branches or metulae 12 to 14 $\mu$  long; sterigmata about 8 $\mu$  in length; conidia about 2 to 2.5 $\mu$  in diameter, globose, with connectives evident, persisting in chains in fluid mounts.

Culture no. 5010.22 received as type from Baarn in July, 1928, appears to satisfy Zaleski's description. The well-established but diverging columns of conidia arising from the branches or metulae ally this species with *P. corylophilum* and *P. citrinum* although certain of the Radiata approach this same picture.

Our no. 5020 isolated in December, 1928, from Collington loam duplicates Zaleski's species very nearly.

136. *P. citrinum* Thom. Emended from U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 61-63, fig. 22. 1910.

Colonies grown upon gelatin and potato or bean agar blue-green when young, becoming dark brown when old, with colored fruit borne almost to the very margin of the colony, so that the white border of submerged mycelium and uncolored fruit is very narrow; restricted in growth to a few millimeters in diameter upon gelatin, but becoming much larger upon agar; aerial part of colony consisting of densely standing conidiophores and conidia except in the center, where there arise a few tufts of trailing aerial hyphae; reverse of colony itself colorless or only yellowish; conidiophores arising separately, rarely longer than 150 $\mu$ , branching

acropetally from submerged hyphae radiating from the center of the colony, or branched from the hyphae of the central aerial tuft; penicillus a verticil 2 to 5 branches 16 to 30 by  $3\mu$  enlarged at apex to  $5\mu$ , each producing a compact verticil of sterigmata bearing chains of conidia massed together into columns 50 to  $150\mu$  in length (usually 80 to  $100\mu$ ).

The whole fructification appears in this way double, triple, or quadruple or even more complex by a secondary verticil from the central branch. Sterigmata 6 to 7 by 2 to  $3\mu$ . Conidia globose when ripe,  $2.4$  to  $3\mu$  (even  $3.5\mu$  diam. in cane-sugar cultures) in diameter, bluish-green, slightly granular in contents, adhering in chains in fluid mounts, losing vitality rapidly with change of color in old colonies. Colonies liquefy gelatin rapidly, so that they lie in pools of liquid within a week.



FIG. 34. *P. citrinum* Thom: a, branched type of colony upon rich substrata; b, typical colony.

Litmus reaction in plain gelatin, strongly alkaline. Produces a lemon-yellow color soluble in alcohol in media containing sugars, milk, gelatin, bread, and potato.

Found in cultures from milk and cheese, probably cosmopolitan.

137. *P. auriflumum* Biourge. La Cellule 33: fasc. 1, pp. 250-252; Col. Pl. VII, Cart. 53; Pl. XI, fig. 64. 1923.

Synonym: *P. citrinum* Thom, which see.

Colonies on wort gelatine restricted in growth, quickly liquefying the gelatine to an orange fluid, blue green, then dark green, coremia none; reverse greenish yellow, dark brick red, fulvous, or colorless; gelatine liquefied; conidiophores about 40 to 50 by  $3\mu$ ; penicillus 25 to  $40\mu$  in

length with all walls smooth; metulae 10 to 13 by 2.5 to 3.5 $\mu$  with apex enlarged to about 6 $\mu$ , in twos, threes, or fours; sterigmata 6.5 to 9 by 3 to 3.5 $\mu$ , in fives, sevens or more; conidia subglobose 2.5 to 3, to 4.5 by 3 to 3.5 $\mu$ .

Biourge's type no. 53 (our no. 4733.14) was purified from a culture obtained from Kral's Laboratory as *P. citrinum*. Biourge failed to observe the cylindrical columns of conidia described by Thom, hence redescribed the species. His culture (4733.14) when grown on Czapek's solution agar, and upon 15 per cent gelatine in water, proved to be Thom's organism and to form the columns of conidia described.

Thom sent his type culture to Kral hence it is not surprising that Biourge found it in Kral's impure culture. Biourge however seems to have accepted Westling's conception of *P. citrinum* applied the name in his own collection to an entirely different organism then redescribed Thom's own type culture as *P. aurifluum*. Our recent notes on Biourge's culture follow: colonies of Biourge's strain no. 4733.14 upon Czapek's solution agar, velvety slowly spreading up to 35 mm. in diameter and 200 to 300 $\mu$  deep in fourteen days, plane, radiately wrinkled and sometimes more or less zonate in the central area, less at margin, margin narrow about 1 mm. white, passing through a narrow bluish green zone to shades of olive green to deep olive gray; reverse uncolored to yellow to pale flesh color which sometimes differs widely as to the agar; drops colorless, rather few central; odor none; conidiophores short; penicillus sometimes a single columnar mass mostly a main axis with a partial verticil of 1 to several more or less divaricate metulae each producing a columnar mass of conidia varying in length from about 100 to 200 $\mu$  with the conditions of culture; metulae about 16 $\mu$  long enlarging from base of 2 to 2.5 to apex more or less vesicular 4 to 5 $\mu$ ; sterigmata about 8 by 2 to 2.5 $\mu$ ; conidia 2.3 to 3 $\mu$  commonly about 2.5 $\mu$ , very pale in color, smooth and homogeneous in apparent contents.

So many cultures have been received with the general morphology of *P. citrinum* that we have felt justified in giving both Biourge's diagnosis and our own notes from recent Czapek cultures. Among the strains examined the following seem worthy of separate notice: no. 4482 isolated by Dr. Frank Forry from sputum of a woman with lung disease; no. 4207.6 from cranberries grown in the Rocky Mountain states; no. 4658.39/9 from fruit, Cape Town, South Africa; no. 4202.17 contributed by Prof. Thaxter as brought from China by Dr. Chung; no. 4725.-805 from C. G. Hansford in Jamaica; no. 4279J from Saito, Dairen, Manchuria; no. 4716.15 from sausage, Washington; no. 4684B from

Mr. McWhorter in the Philippines. Although it is entirely possible that careful biochemical studies would separate some of these forms, they belong in a series in so far as routine culture is adequate for grouping. Repetition of this morphology from widely separate regions is fairly good evidence of a fairly well-established series.

*Sub-section 2. Radiata Biourge, La Cellule 33, fasc. 1, pp. 167-168.*  
Type species *P. chrysogenum* Thom.

Sub-section diagnosis: Colonies azonate or at most faintly zonate in age, velvety to lanose in appearance, in some showing a basal network of aerial hyphae, called Radiata by Biourge from their regular contour and the consistently radiating submerged hyphae; penicillus variously combining the complex branching system of the asymmetrica with the retention of the individuality of each metula with its verticil of sterigmata and its column of conidial chains, especially showing the appearances of monoverticillate penicilli in accessory branches below the terminal verticil of metulae, these branches irregularly spaced, unequal in length and variously divergent; penicilli in the same colony often partly complex masses, partly divergent and suggesting a series of branches each producing a monoverticillate mass (fig. 35).

Biourge discussed this as a natural group for which he took *P. griseo-roseum* Dierckx (1901) as the type and cited *P. notatum* and *P. chrysogenum* as the well known representatives. He added *P. citreo-roseum* Dierckx (incorrectly listed by Dierckx as monoverticillate), also *P. rubrum* of Grassberger and Stoll on the basis of a culture from Kral, also *P. brunneo-rubrum* Dierckx, *P. baculatum* Westling, and his own two species *P. cyaneo-fulvum* and *P. roseo-citream*. Zonation was noted as entirely absent from most of them, and even when present combined with a prominent radiation of hyphae which links the forms with the non-zonate series. Overgrowths are reported to be common in old cultures, and to be in color rose, with outer margins often similar in color.

Biourge's failure to describe the fruiting masses as seen in the growing colony weakens his description throughout. In the Radiata the terminal group of metulae is well established in contrast to the penicillus of the species of divaricate sections, but the monoverticillate fruit mass predominates in the accessory branches. In the terminal verticil, each metula often preserves much of the individuality of a monoverticillate head with a columnar mass of conidia.



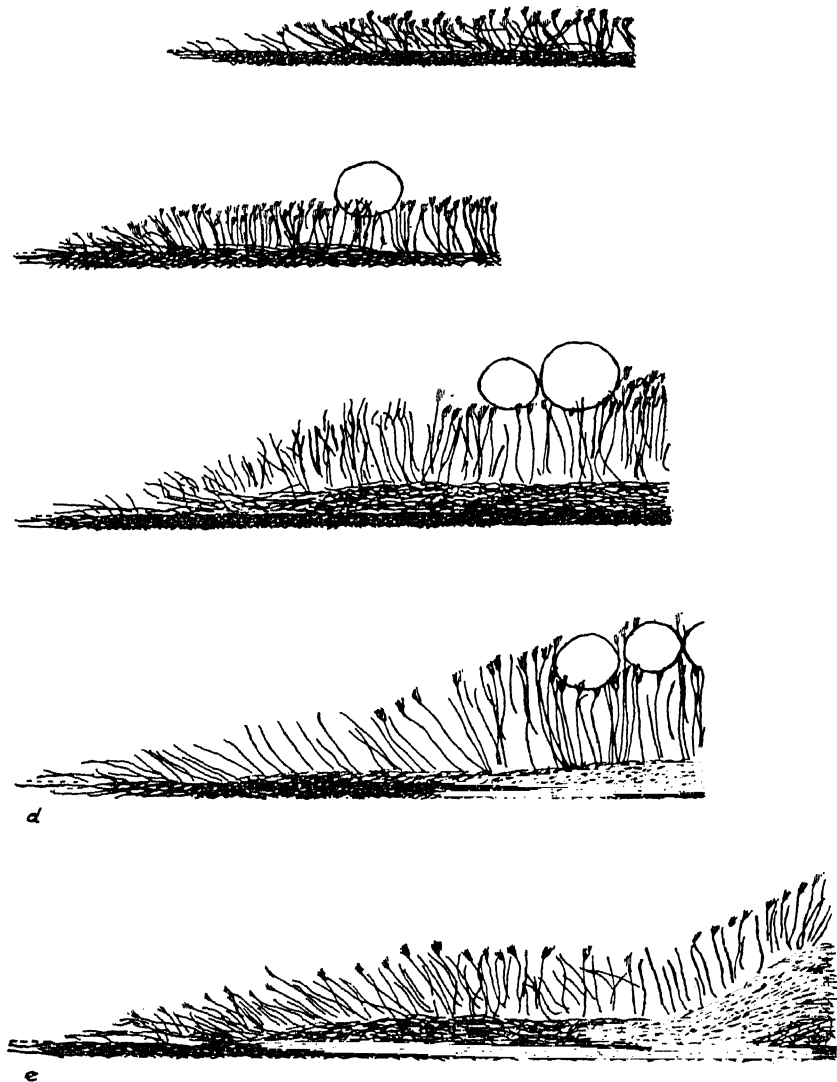


FIG. 35. Series of diagrammatic radial sections of species described in the "Radiata" (magnified 25 times): a, *P. chrysogenum*; b, *P. baculatum*; c, *P. notatum*; d, *P. brunneo-rubrum*; e, *P. cyaneo-fulvum*.

The numbers of sterigmata and metulae in the vesicle as reported by Biourge are too small to represent the conditions we have seen. Such numbers as 2 to 5 are far too low for the usual penicillus, although here as always, occasional fruiting masses show small numbers of elements.

Great difficulty will be encountered in the allocation of the members of the *P. chrysogenum* series to the various species proposed as cited here. After one of them is known little difficulty is met in recognizing most of these forms as members of this series. The arrangement proposed recognizes the superficial differences observed in culture.

Some allocations may appear arbitrary. In the color and superficial appearance of its velvety colonies *P. griseo-brunneum* Dierckx in some cultures appears to belong with the *Radiata* but its penicillus with tangled conidial chains have lead us to place it with "*Brevicompacta*."

140. *P. chrysogenum* Thom. In U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 58-60, fig. 20, 1910; see also Biourge Monogr. La Cellule 33, fasc. 1, pp. 170-172, Col. Pl. IV, Cart. 163; Pl. VI, fig. 32, 1923; and Westling, Arkiv f. Bot. 11: 54, 107-108, figs. 23 and 64. 1911; type culture Thom's no. 26.

Colonies on Czapek's solution agar azonate velvety or with basal scattered aerial hyphae, in age more or less overgrown with white or rosy tufts of hyphae, especially in central areas, broadly spreading with white marginal area about 2 mm. wide in the growing period, and conidial areas greenish glaucous blue (Ridgway XLII) then green, becoming some shade of violaceous-brown in age, or almost fuscous upon other media, the mass about 300 to 400 $\mu$  deep; with reverse in yellow tints in Czapek; drops abundant uncolored or yellow; odor none; conidiophores mostly arising separately from submerged hyphae about 300 by 4 $\mu$  commonly also as longer, branching and ascending fertile hyphae at the margin in older colonies, penicillus usually a terminal verticil of metulae and various accessory and more or less diverging branches below, with verticils of sterigmata commonly suggesting monoverticillate penicilli and producing mostly columnar masses of conidia; metulae 10 to 16 by 3.5 to 4 $\mu$ ; sterigmata 8 to 9 by 3.5 $\mu$ ; conidia from elliptical 4 by 3.3 to subglobose about 4 $\mu$  mostly when ripe.

Species digests milk rapidly producing a clear golden yellow liquid, liquefies gelatin rapidly with yellow color and alkaline reaction; colors potato yellow, but produces no color on potato-agar or bean agar without added carbohydrate.

No. 4733.33 *P. chrysogenum* from Biourge in Czapek's solution agar

showed an abundance of rather small yellow drops; yellow below and diffused into the agar.

Literally hundreds of the cultures examined have shown the general characters of *P. chrysogenum*. These differ in shades of blue green, green, and even yellow green, in rate of spreading, in depth of aerial mass which varies from strictly velvety about  $200\mu$  to lanose or almost floccose  $1000\mu$  or more. In morphology of penicillus they vary from strains in which the separateness of the individual verticil gives the effect of a fertile hypha bearing a series of branches each with a monoverticillate conidial mass, to a complex typically asymmetrical conidial apparatus. The conidia vary from quite small as in *P. notatum* with the diameter mostly less than  $3\mu$  to forms with conidia  $4, 4.5\mu$  or more in long axis and more or less definitely elliptical.

No list of cultures examined will be given. They came from every region from which either cultures or materials have been studied. They grow readily under widely different conditions from cool to blood heat ( $37^{\circ}\text{C}.$ ). One culture remained viable eight years in a laboratory test tube. They differ in gross appearance when side by side in culture, but when great numbers of them are examined the definiteness of distinction slips away and blends in a picture of a series with its range of reactions.

Studied upon a series of culture media, *P. chrysogenum* (no. 26) changes color with the composition of the substratum and with the age or stage of metabolism of the colony itself; colonies grown on Czapek's solution in the presence of abundance of fermentable sugar long remain green and often reach a pH of 4 to 3.6, without such sugar little or no yellow appears in the substratum and the conidial areas turn dark almost fuscous in age.

Re-examination of records of observations over the twenty-two years this species has been kept in culture shows as great a range in color in the various recorded experiments as is shown by the whole series of Radiata. There thus appears to be a group with real relationship within which quantitative variations occur definitely enough, perhaps, to justify the establishment of Biourge's series of species names. These names may, however, be expected to mark out contrasting sections of a great series of intergrading strains rather than to offer to the worker assurance from his own identification that he has rediscovered the exact organism described by either of us.

141. *P. chlorophaeum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 271-273; Col. Pl. VIII, Cart. 39; Pl. XIII, fig. 78. 1923.

Colonies on wort gelatine, more or less restricted in growth, wrinkled,

velvety, at first green (viridis) then darkening toward cinereous and finally brown; coremia none; reverse pale sordid rosy to yellow brown; odor mostly none; conidiophore 30 to 90 by  $2.4\mu$ , with all walls smooth; penicillus a simple verticil of sterigmata about  $10\mu$  long, or a verticil of metulae up to  $30\mu$  long; metulae 13 to 20 by  $2.4$  at base and  $4\mu$  above, enlarging toward the apex; sterigmata 7 to 12 by  $2.5$  to  $4\mu$ , three to many in the verticil; conidia 2.8 to 3.2 by  $2.5\mu$  to subglobose  $3.6$  to  $4.2$  by  $3$  to  $4\mu$ .

Biourge's type no. 39 (our no. 4733.31) when grown on Czapek's solution agar produced colonies velvety with a felt of hyphae near the agar but rising slightly above it, azonate, radiately wrinkled, becoming up to  $600\mu$  deep; with broad white margin; blue green passing to gray green to shades of gray or purplish gray in age; reverse and agar at first yellow then through orange shades brown, with yellow more or less persistent at margin; drops abundant colorless; odor none. Colonies digest milk and color the liquid yellow.

Several cultures have been seen with the color changes described for this species which seems to indicate that the characters indicated are frequently reproduced. Nos. 5037.107 and 122 from the British Cotton Industry Research Association produced colonies blue green to green in age dark shades toward slate-olive, deeply and loosely velvety or almost lanose; reverse and agar yellow toward orange or reddish orange to deep brown shades; penicilli with elements either in diverging columns or forming a single mass up to  $400\mu$  long. Conidia are elliptical to subglobose about  $4\mu$  in long axis.

2. *P. griseo-roseum* Dierckx. Soc. Scientifique Bruxelles 25: p. 89. 1901. In Biourge, Monogr. La Cellule 33: fasc. 1, pp. 168-170; Col. Pl. IV, Cart. 29; Pl. VI, fig. 31. 1923.

Colonies on wortgelatine, velvety, with broad white margin and conidial areas blue green (C.d.C. 361-362), in age overgrown with rosy mycelium; becoming wrinkled and buckled, with the liquefaction of the gelatine; reverse at first uncolored, then yellow, in age; (in Czapek's solution agar, colonies velvety up to about  $500\mu$  deep with radiating creeping partly submerged partly aerial hyphae, and in age overgrowth of mycelium in the center, with white margin of the growing colony about 3 mm. broad with superficial creeping hyphae to the very edge); conidial areas blue green to green with abundant but loose mealy masses (not crusts) of conidia, reverse and agar pale yellow; drops abundant, large, colorless; conidiophore about  $4\mu$  in diameter, with all walls smooth

(in Czapek up to 350 to 500 $\mu$  in length); penicillus about 60 $\mu$  long, figured, as a main axis with a terminal verticil of metulae with paired or single branches either monoverticillate or biverticillate, short or long, commonly more or less diverging at 1 to several nodes of the main axis; branches paired 20 to 30 $\mu$  long; metulae mostly 10 to 12 or even 16 $\mu$  by 3 $\mu$ , occasionally very much longer; sterigmata 7 to 10 by 3 $\mu$  in verticils of 3 to 5 (or 7?); conidia globose 3 $\mu$  or elliptical 3.5 by 4 $\mu$ .

Biourge's type no. 29 (our no. 4733.70) shows the characters of the group, digests milk producing a golden yellow fluid, and produces some of the rosy areas emphasized in the name when grown in gelatine.

*P. roseo-griseum* probably a misprint for *P. griseo-roseum*. In Biourge, Monogr. La Cellule 33: 171, 1923.

143. "*P. griseo-rubrum* Dierckx" is found on Colored Plate IV of Biourge for his no. 29 which in the text is made the type of *P. griseo-roseum* Dierckx. This is probably a misprint since the name is not referred to elsewhere.

144. *P. notatum* Westling. Arkiv för Botanik 11: 55, 95-97; fig. 17, 59. 1911. Compare Biourge, Monogr. La Cellule 33: fasc. 1, pp. 179-181; Col. Pl. IV, Cart. 19; Pl. VIII, fig. 37. 1923.

Colonies in prune gelatine, floccose, blue green (C.d.C. 397, 388, 383) with broad white margin; reverse subflavus; gelatine quickly liquefied, with an acid reaction; vegetative hyphae commonly 5 $\mu$  in diameter; odor often detectable but weak; conidiophores smooth, mostly from submerged hyphae, partly as branches from aerial hyphae, up to 750 $\mu$  by 2.8 to 4.6 $\mu$ , occasionally branched toward the base, penicillus 45 to 135 $\mu$  long, figured as a verticil of 3 metulae with their groups of sterigmata and conidial chains, in Westling's culture no. 2541 the morphology approaches that of *P. chrysogenum*; metulae 10.5 to 14 by 3 to 4.6 $\mu$ ; conidia smooth subglobose 2.6 to 3.2 $\mu$ , enlarging to 5.2 to 6 $\mu$  in germination.

Our no. 2541 from Westling appears to be type.

Found on rotting branches of Hyssopus, in Norway; also under the name *P. glaucum* from the Centralstelle für Pilzkulturen. It grew well upon Westling's media, in milk produced mycelium yellow in reverse and a yellow color in the digested milk; colonies grew well at 30 to 31°C. Biourge apparently obtained the same culture (his no. 19) from Kral in 1913 and it appears in our collection as 4733.90.

The type, our no. 2541, received from Westling appeared to be identical with our no. 102 received from Amsterdam, both belong in the group with *P. chrysogenum* and show globose spores somewhat smaller than the type. Organisms close to *P. notatum* have been repeatedly found.

145. *P. virescens* Bainier. Bul. Soc. Mycol. France 23: 12, pl. II, fig. 9-12. 1907. Probably near *P. notatum* Westling. not *P. notatum* Westling. Not *P. virescens* Sopp, which see.

Colonies on licorice sticks, green to blue green, or grayish blue green, described as giving a powdery look to the substratum suggestive of *P. digitatum*, hence probably spreading, velvety; submerged hyphae slightly undulate, radiating, slender; "aerial hyphae" (conidiophores apparently) little developed, with many divergent branches fairly close together, varying considerably in length, each ending in a penicillus, which may vary from a simple verticil of sterigmata, to a verticil of metulae bearing sterigmata or to a primary series of branches preceding the metulae; sterigmata about  $8.4\mu$  long; conidia about  $2.8\mu$  in diameter, swelling very greatly in germination and producing 1 or rarely 2 tubes.

Bainier's species has not surely been recognized by any succeeding worker. Our no. 4640.457 under this name from the Bainier collection does not satisfy this description. Compare our no. 24 in Bul. 118 which satisfies the description except for the comparison to *P. digitatum*, which calls for a broadly spreading form and taken with Bainier's figures probably puts this in the *P. chrysogenum* series and with the members of that series showing small conidia  $2.8\mu$ ! hence suggesting *P. notatum* Westling.

Bainier's descriptions from colonies grown upon licorice sticks lose the characteristic yellow colors produced by this series of forms in most media. Without these striking colors his failure to separate them satisfactorily is easily understood.

Sopp used this species name in 1912 but certainly for a different form.

146. *P. citreo-roseum* Dierckx. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 182-184; Col. Pl. IV, Cart. 18; Pl. VII, fig. 38. 1923.

Colonies on wort gelatine azonate beautiful sky blue, then olive green, finally gray rose, to reddish brown with rosy tomentum (overgrowth?); coremia none; reverse orange, then red (rose-carmine), drops at first colorless then red; odor none; conidiophore 3 to  $3.5\mu$  in diameter; penicillus 35 to  $50\mu$  long with all walls smooth, figured as a central axis with or

without one or two appressed branches from the uppermost node, then closely packed metulae and sterigmata at a single level; branches up to 15 to 25 by  $2.5$  to  $3.5\mu$ , in pairs or threes; metulae 10 to 12 by 2 to  $2.6\mu$ , in twos, threes, or fours; sterigmata mostly 7.5 to 8 by  $2.4\mu$ , in twos or threes; conidia at first 2.8 by  $2.5\mu$ , then subglobose 3 to  $3.5\mu$ .

Type: Biourge's no. 18 (our no. 4733.36) is recorded as having been furnished by Kral to Dierckx as *P. roseum*. In cultures on Czapek's solution agar, tomentose to  $1000\mu$  or more in depth, or with a felted base supporting a tangled tomentum, velvety or nearly so in the marginal area, slowly bluish green to gray green, Ridgway's deep glaucous gray (XLVIII), becoming dark gray in age; reverse and agar yellow then slowly red in center; drops abundant uncolored; conidiophores very slender 2.5 to  $3\mu$  in diameter and often  $1000\mu$  in length prostrate or ascending tomentose or matted not forming a vertically crowded area; penicillus variously a main axis and verticil of diverging metulae, or axis and one or more branches more or less diverging and bearing either sterigmata or metulae.

147. *P. meleagrinum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 147-149; Col. Pl. III, Cart. 124; Pl. IV, fig. 22. 1923.

Colonies on wort gelatine semifloccose to tomentose-lanose, wrinkled and buckled, conidial areas blue (C.d.C. 422, 397, 392), then blue green (C.d.C. 362), with marginal white zone 1 mm. broad, zonation indicated by placing but not described; coremia none; reverse colorless or pale yellow; odor none or indefinite; conidiophore about  $3\mu$  in diameter, with wall smooth; penicillus about  $40\mu$  long (20 to  $25\mu$  when branches are wanting), figured as main stalk and 1 branch, unequal, more or less divergent, bearing compact verticils of metulae and rather coarse sterigmata but in his sketch several diverging branches at various levels are suggested; branches 2 or 3 or none; metulae 7 to 14, commonly 11 to 12 by 2 to  $3\mu$ , enlarging toward the apex, in verticils of 2 to 4; sterigmata 7 to 9 by 3 to  $3.5\mu$ , in groups of 2 to 4; conidia 4.5 by 3.2 to  $3.6\mu$ , scarcely deciduous.

Biourge no. 124 was not received by us but was described as characterized by its aerial down (duvet), its reverse spotted (guinea hen appearance), and the blue of its young conidia; in its growth it was a border culture between the stellate and radiate series.

A culture, no. 5034.53, received in January, 1929, from Nobel's Explosions Company, through J. H. Birkinshaw seems to satisfy Biourge's diagnosis well enough to be assigned there; Colonies grown upon Czapek's solution agar velvety spreading azonate, plane or with

radiate wrinkles and a central elevation or umbo, with white margin 1 to 3 mm. wide, a band of pale blue green about 5 mm. wide within passing to bluish gray green or deep dark gray green in age; reverse colorless to reddish, violaceous or drab shades; drops when seen crystal; odor faint "fruity"; conidiophores 150 to 200 by 3 to  $4\mu$ , with walls smooth; penicilli consisting of main axis and 3, 4 or 5 unequal metulae producing either a single columnar mass of conidia about  $100\mu$  long or a column for each metula; metulae 10 to  $12\mu$  long; sterigmata 8 to  $9\mu$ , with young conidia showing a distinctly cylindrical phase; conidia elliptical to subglobose 3, 3.5 or  $4\mu$  in long axis.

148. *P. cyaneo-fulvum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 174-176; Col. Pl. IV, Cart. 128; Pl. VI, fig. 34. 1923.

Colonies on wort gelatine, at first sky blue (azureus) (C.d.C. 403<sup>b</sup>), then darkening, finally rose amethyst (rose purple) (C.d.C. 99, 119), with wide white margin, reverse dull yellowish; coremia none; conidiophore 2.5 to  $3.5\mu$  in diameter, with all walls smooth; penicillus about  $50\mu$ , figured as a main axis with its terminal verticil of metulae with irregular branch, branches 20 to  $30\mu$  long in pairs or threes; metulae 8 to 13 by 2.5 to  $3\mu$ , in threes or fours; sterigmata 8 to 10 by 3 ( $3.5\mu$  ?), in verticils of 2 to 5 (in ours 8 by  $2\mu$ ); conidia globose 3.5 to  $4\mu$  or ovate 4 to 5 by  $3.5$  to  $4\mu$ , caducous.

Biourge's type culture no. 128 (our no. 4733.47), grown on Czapek's solution agar, produced colonies velvety with submerged margin, a broad white zone from  $100\mu$  to 300,  $400\mu$  or even  $500\mu$  deep toward the center, where the mass becomes more or less wrinkled and up to 1500 in depth, bluish gray green, reverse and agar yellow, to reddish or coral pink (Ridgway XIII), with rich yellow drops, hyphae more slender than other members of the series.

Colonies digested milk rapidly with bright yellow color.

149. *P. brunneo-rubrum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88, 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 176-179; Col. Pl. IV, Cart. 48; Pl. VI, fig. 36. 1923.

Colonies on wort and Raulin solution gelatine zonate described and figured as zones separate, sky blue to olive green, with broad white marginal zone, coremia none; reverse variegated yellow, greenish, or with fimbriate appearance outer tips rose, then yellow and green; gelatine liquefied and yellowed; drops yellow with a greenish tinge, odor none; conidiophore about  $3\mu$  in diameter; penicillus 40 to  $70\mu$  long, all walls



smooth, figured as main axis tipped by a verticil of sterigmata on a central metula surrounded by a loose verticil of a few metulae, and often with one or more fairly long diverging branches from nodes below and with secondary penicilli from monoverticillate to biverticillate; branches none or alternating, 25 to 60 by  $3\mu$ ; metulae 10 to 20 by 3 to  $3.2\mu$  in verticils of 2 to 4; sterigmata 10 to 15 by  $3.2$  to  $3.5\mu$  in verticils of 4 to 7; conidia elliptical  $3.5$  to  $5$  by  $2.5$  to  $4$ .

Biourge type no. 48 (our no. 4733.22); in milk produced a pale yellow rather than the golden digest liquor so characteristic of the *P. chrysogenum* group although the reverse of the colony showed some yellow; in Czapek's solution agar, colonies faintly zonate, velvety, loose, not crowded, up to 1 mm. in depth, bluish gray green, then sage green, finally brown, with broad white margin during the growing period; reverse and agar yellow in zones, becoming reddish or reddish brown in center (shade of red or rose varies with the medium), drops often colorless or sometimes pale yellow.

150. *P. baculatum* Westling. (Westling, R. En my ascusbildande Penicillium-art. Svensk Botanisk Tidskrift 4: 139-145, text figures 1-3, 1910.) See also Arkiv för Botanik 11, pp. 53, 79-83, fig. 11, 53. 1911. Compare Biourge Monogr. La Cellule 33: fasc. 1, pp. 186-188; Col. Pl. IV, Cart. 384; Pl. VII, fig. 40. 1923.

Colonies in prune gelatine, floccose, white then bluish green (C.d.C. 366 to 388, 389, 393) with a white floccose margin; gelatine rather slowly liquefied and alkaline; sessile hyphae colorless, 2 to  $5\mu$  in diameter, and among them in the substratum needles or columnar crystals of calcium oxalate, mostly in bundles or packet; odor almost wanting; conidiophores with walls smooth, 50 to  $800\mu$  long by  $3.4$  to  $5\mu$ ; penicillus figured as a terminal verticil of metulae; metulae 10 to 14 by  $4.2$  to  $6\mu$ ; sterigmata  $6.5$  to  $9\mu$  by 3 to  $3.4\mu$ ; conidia smooth, oval to elliptical,  $3.8$  to  $4.6$  by 3 to  $3.6\mu$ , in germination up to  $5.3$  or even  $7.5\mu$ ; perithecia (rarely produced ? see note below) yellow, thin-walled; asci hyaline, globose to ovate; ascospores  $4.2$  to  $4.8$  by  $5.2$  to  $6\mu$  lenticular (2-valved) with edge subcanaliculate (= with a shallow furrow), as described and as the ascospores were figured these perithecia belong in *Aspergillus*.

Species found upon leaves in Norway. Cultures grew well at  $30$  to  $31^\circ$ , produced mycelium but no spores at  $37^\circ\text{C}$ . Colonies grew well on all Westling's media, in milk producing a rich yellow color.

Two cultures were received from Westling under this name, 2550, and 2550a, both of which clearly belong in the group with *P. chrysogenum*

Thom. The first (no. 2550) failed to produce perithecia. After Westling found his second tube containing perithecia, he sent another transfer no. 2550a from which many transfers failed to produce perithecia but a mount, showed a suspicious structure. The addition of sugar in high percentage was followed promptly by the development of a perithecium producing member of the *A. repens* section of the *Aspergillus glaucus* group.<sup>1</sup> In spite of Westling's belief in the purity of the culture, there is good reason to believe that the perithecia studied and described by him belonged to the *Aspergillus* which grew in this experiment. Our no. 2550a was sent to Biourge and later returned by him as his no. 384 (our no. 4733.16).

Westling's organism appears to be more nearly floccose than other members of the series and to produce conidia with more pronounced ellipticity. The following notes from our own culture may be added.

Biourge's culture no. 384 *P. baculatum* (our no. 4733.16) was sent to Biourge by us as no. 2550a and later returned under the above number. He regards the conidia as much more elliptical than in *P. chrysogenum*. Our observations follow: Colonies of no. 4733.16 in Czapek's solution agar, velvety 400 to 600 $\mu$  in depth, spreading broadly, green (American green, see Ridgway XLI.33''') with blue shades; with hyphae fairly coarse, reverse colorless at first, slowly yellowish to brownish rather than bright yellow; drops abundant; more or less yellow or citrine conidiophores smooth, more or less sinuate, branching variously and divergently toward the apex; metulae in compact verticils or absent and replaced by diverging branchelets; sterigmata 7 to 10 by 2 to 2.5 $\mu$ ; conidial elliptical to globose 4 $\mu$  or slightly more in long axis.

When grown in milk it produces a yellow digested fluid under a thin wrinkled mass of mycelium.

*Sub-section 3. Velutina-restricta:* Colonies velvety, narrow bordered, or restricted in colony growth habit; zonation sometimes showing in marginal areas of old colonies ("hemizonate" of Biourge), conidiophore walls more or less pitted, granular or roughened; series typified by *P. puberulum* Bainier.

Compare sub-section 5, in which the broadly spreading habit of growth is the principal difference.

There are a number of organisms intermediate in habit between the Radiata and Stellata of Biourge's monograph. They are distinctly

<sup>1</sup> See The Aspergilli by Thom and Church.

velvety in appearance, narrow bordered, restricted in growth, azonate or hemizonate (Biourge) not definitely zonate, and show the punctate-pitted (or "rough") wall which is so characteristic of several sections and subsections in *Penicillium*. Lines of separation are so difficult to define that most of the descriptions if unaccompanied by verifiable cultures are uninterpretable. Certain described forms have been brought together in this subsection as centers about which cultures may be grouped and foundations laid for a real comparative understanding of these species.

155. *P. casei* Staub. Centralb. f. Bakt. 2 Abt. 31: 454-466. 1911.

Probable synonym: *P. biourgei* Dierckx.

Colonies on neutral whey gelatine at first white, then powdery gray green; mycelium a tough close woven felt partly aerial but with crowded conidiophores appearing almost velvety; reverse clear yellow to brownish then dark brown; on milk agar especially marked brown color; gelatine liquefied, with alkaline reaction; conidiophores 80 to 150 $\mu$  by 3 to 4 $\mu$ , with walls granular, penicillus consisting usually of central stalk and branch or branches rather long appressed, bearing verticils of metulae 10 to 15 $\mu$  long; sterigmata 7.5 to 10 $\mu$ , few in the verticil; conidia smooth, elliptical 3 by 2 at first, then globose about 3.5 $\mu$ .

Species causing brown spots in the rind of Swiss (Emmentaler) cheese. Staub's culture (our no. 2756) was received and studied but lost. Similarly other strains were isolated from such spots on Swiss cheese and lost. Our notes from culture follow: Colonies upon Czapek's solution agar, forming thin tough close textured felts, buckled and wrinkled, in bluish glaucous to dull green shades, 150 to 250 $\mu$  deep; reverse yellow to orange brown to deep brown almost black, otherwise as described by Staub. A stock culture carried as "*P. casei* D" has maintained the distinctive characters above for many years. It was isolated by us from Swiss cheese which showed the spotting described by Staub.

156. *P. Biourgei* Dierckx. Soc. Scientifique Bruxelles 25: p. 88. 1901.

Spores 3 to 4 $\mu$  assez persistantes. Sterigm. 3 to 10. Fructif. 60 to 160 $\mu$  a rameaux assez divergents. Spores glauque fonce passant au brun sombre avec taches blanches. Revers incolore, rose, brunatre. Sur fromage diffusion de noir par places.

Biourge Monogr. La Cellule 33: fasc. 1, p. 167, 1923, placed this species provisionally in his "hemizonata, category inflata" and says he has seen the black spots made by it in cheese upon Dierckx's table but has not been able to identify it in his cultures.

157. *P. puberulum* Bainier, Bul. Soc. Mycol. France 23: 16-17; Pl. IV, fig. 6-12. 1907. See fig. 36, 37.

Colonies upon licorice sticks, vigorous, blue green, becoming dark blue green in age; conidiophores (length not given but noted only as "plus ou moins allongé") about  $5.6\mu$  in diameter, figured as somewhat sinuous, not always smooth, at times with walls showing "fine granulations" difficultly visible, commonly also many vacuoles, occasionally bearing a long branch toward the base; penicillus figured and described

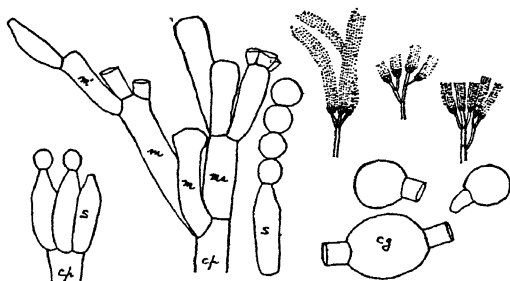


FIG. 36. *P. puberulum* Bainier (Alsberg and Black strain): *cp*, apex of conidiophore; *ma*, main axis extended; *m*, metulae; *m'*, secondary metula from proliferation of one of them; *cg*, germinating conidia and habit diagrams of penicilli.

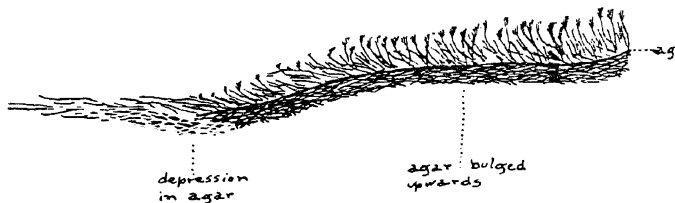


FIG. 37. *P. puberulum*: Diagrammatic radial section of colony

as usually sparingly branched with rather long more or less diverging primary branches, bearing metulae or secondary branches or both in the same verticil; sterigmata not described; conidia globose, more or less unequal in diameter but averaging  $4.2\mu$ , swelling slightly before germinating and emitting one or more germ tubes.

The culture received from the Bainier collection under this name (4640.454) corresponds with Bainier's description of *P. patulum*, hence must be rejected.

Shortly after the publication of Bainier's description a culture isolated by F. D. Heald from rotting corn (*Zea Mays*) at Lincoln, Nebraska, was submitted for study and ultimately transferred to Alsberg and Black of the Bureau of Plant Industry for use in biochemical work. When their work was published they asked for an identification of this culture which had been carried as Heald's 45 (H. 45). Since it appeared to correspond fairly closely to Bainier's description a tentative identification as *P. puberulum* Bainier was given and a description with figures from our own cultures was added which appears on pp. 12-13 in United States Department of Agriculture, Bureau of Plant Industry, Bul. 270, 1913. This culture was sent by them to Dr. Westerdijk as *P. puberulum* Bainier. In the meantime we lost our culture. In 1926, Dr. Westerdijk returned this culture to us (4876.20) as "*P. puberulum* Bainier, Alsberg and Black." It has been checked against the description and found satisfactory. Since this culture appears to be verifiable as the culture which furnished the penicillic acid discussed by Alsberg and Black as produced by *P. puberulum*, an emended form of the description furnished to and published by them is given here:

Colonies on Czapek's solution agar velvety, plane or somewhat raised occasionally almost unbonate in the central areas, but not radiately wrinkled, restrictedly growing, azonate during the rapidly growing period, developing a broadly spreading very thin zonate area covering the whole surface of the medium after several weeks, dense in central areas, bluish green, then green (Ridgway Pl. XLII), persistently green after several weeks; with aerial hyphae following the submerged margin closely at first, but later showing a very broad, thin, radiating submerged area with thinly scattered zonately arranged penicilli; reverse yellowish or greenish to tan; agar uncolored; odor moldy to sourish, rather strong; conidiophores 100 to 200 by 3.5 to 4 $\mu$ , slightly sinuous, with walls variously smooth or more or less pitted or roughened; penicillus consisting of one composite fairly dense column or several more or less ragged and slightly divergent columnar masses, which form fairly dense crusts of hyphae breaking off in masses in old cultures, with branching system consisting of a terminal verticil of metulae with branching from a lower node, all elements enlarged and more or less vesicle-like at the apex; sterigmata 7 to 10 by 2 to 2.5 $\mu$ ; conidia 3.5 to 4 $\mu$  at first elliptical then quickly globose, swelling in germination to 5 to 8 $\mu$  and emitting one or two germ tubes.

Culture no. 45 liquefied gelatine in distilled water with a trace of brown color in the liquid, and an alkaline reaction; the conidial area became

dark or smoky in age in media lacking sugar; it produced no acid from lactose; upon rich media colonies were overgrown with white sterile hyphae in age; seeded upon Raulin's solution the floating colonies failed to fuse, but became buckled, wrinkled, and folded discs of mycelium.

In spite of Bainier's expressed belief that *P. puberulum* and *P. asperulum* were closely related, we have continued to use this name for the organism studied by Alsberg and Black and to apply the name *P. asperulum* to certain widely spreading strains discussed later.

158. *P. Melinii* Thom n. sp.

Colonies in Czapek's solution agar, rather slowly growing 25 mm. in diameter in eight days, velvety, plane or slightly raised in the central area with centers more or less raised or sometimes umbonate, greenish gray or with a bluish effect (Ridgway's Hathi gray LII); with margin (fimbria) white 0.5 to 1 mm. wide; reverse and agar citrine at first becoming a shade of orange brown or russet (Ridgway XV) drops yellow, small, abundant over the whole area, later running together and becoming a shade of orange yellow or tawny or russet. (Ridgway XV); conidiophores with walls granular tuberculate, usually less than  $100\mu$  long by 2 to  $3\mu$ ; penicilli terminal and monoverticillate or a terminal group of diverging and unequal branches or metulae 10 to  $20\mu$  by 2 to  $2.5\mu$ , or with branches irregularly developed at lower nodes, at the tip of the main stem and prolonging it by each successive branch outgrowing the older one in length; sterigmata 6 to  $9\mu$  by  $2\mu$ , with beak like tubes, with chains of conidia divergent or tangled; conidia rough tuberculate 3 to  $3.5\mu$  in diameter, with connective evident.

Contributed by Dr. Elias Melin from forest soil (Type no. 5007.85). Melin's no. 86 was the same species with a sterile contaminant. In colony character this species resembles *P. rubens* Biourge, and some of the velvety monoverticillata. It does not appear closely related to the other species assigned here. This species with velvety colonies, rough conidiophores, verticils of metulae and spinulose conidia probably comprises a series of strains which are not otherwise provided for.

*Sub-section 4. Stellata* Biourge. Monogr., pp. 29-30; pp. 199-209. Includes *P. roqueforti* and its allies.

*Sub-section characterization:* Colonies widely spreading, azonate, velvety, with margin broad veil-like or cobwebby and irregular or uneven, and tending to develop a stellate form (during its early growing period), (fig. 38, 39).

Biourge selected the name *Stellata* to emphasize the contrast in form, between the smooth, even or regular margin of the colony of *P. chrysogenum* with its allies and the more or less uneven or irregularly developing colony of *P. roqueforti*, although the name itself, *stellata*, seems to us to exaggerate the appearances actually encountered. Another species represented by Birkinshaw's culture labeled *P. schneegii* is so definitely stellate in appearance that it must be mentioned here as belonging with the *fasciculata* (see no. 330).

Various authors, Sopp, Weideman, Westling, and even Biourge with small numbers of cultures of comparatively few strains before them have described several species in this series of organisms. Undoubtedly the contrasts between the cultures they had in hand seem to justify the separation of these species. The difficulty of allocating the cultures encountered, among these described species, becomes acute when hundreds of them are compared.

Members of this subgroup are the dominant molds found in spoiling ensilage; they appear frequently in miscellaneous cultures from food and



FIG. 38. *P. roqueforti* Thom: Diagrammatic radial section of margin of young and growing colony.

soils; they dominate the biochemical activities found in the whole group of cheeses characterized by streaks or "marbling" of green mold as seen on the cut faces of cheeses in section. Nearly related species or varieties appear in the sheep's milk cheese (Roquefort) of southern France, in the blue cow's milk cheeses (fromages bleus) of France, in Gorgonzola as made in northern Italy, in Stilton as made in England, and in many less known varieties of loose textured cheese which are made in various lands and which obtain their characteristic flavor from the mold which lines the channels and cracks throughout their mass.

Bainier must be assumed to have seen *P. roqueforti*, since in section IX of his "Mycothèque" (Bul. Soc. Mycol. France 23: 9. 1907) he referred to his study of cultures prepared for use in the making of "fromages bleus d'Auvergne." From other sources (Thom, 1906) we know that this was mold powder prepared from bread inoculated with a mold culture more or less pure, and incubated. In his subsequent discussion Bainier failed to specify which if any of the species described were isolated from this powder and nowhere recognized the Roquefort mold

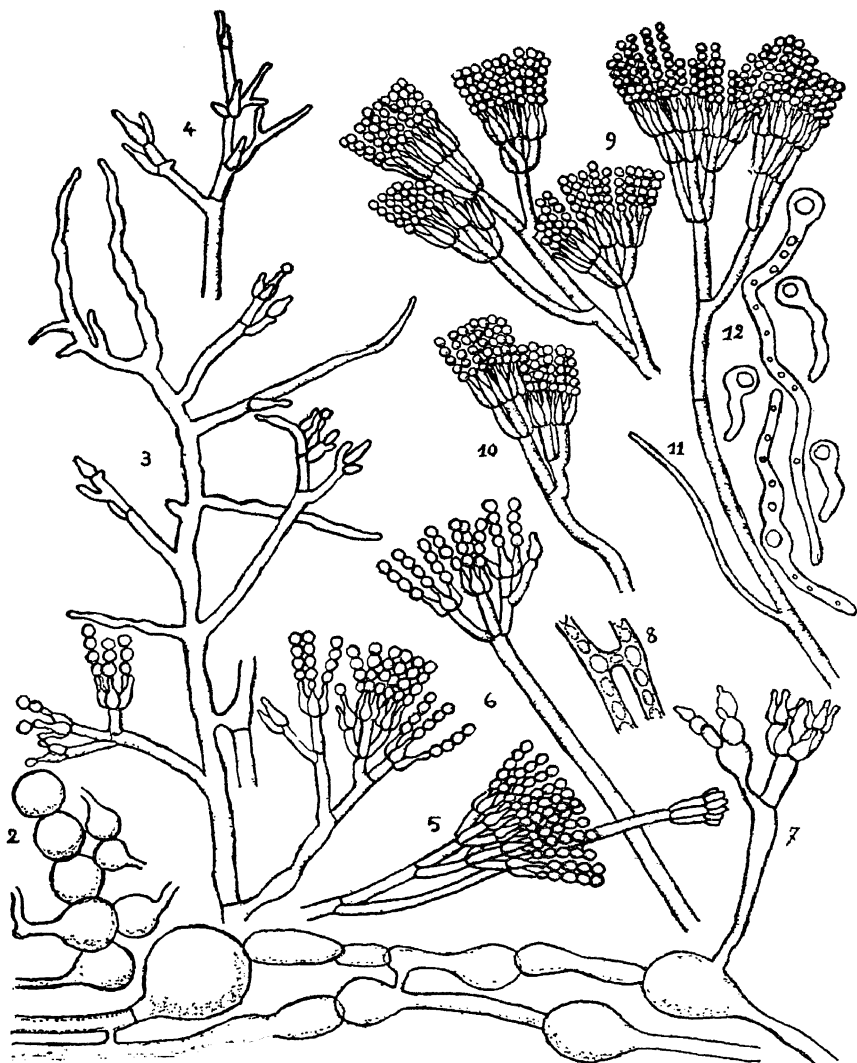


FIG. 39. Bainier's figures of *P. vesiculosum* and *P. virescens*, which are fairly typical of the *P. roqueforti* series: 1, 2, 3, 4 and 7 peculiar and probably pathological cell forms; 5 and 6 fairly typical penicilli; 9-12-*P. virescens* penicilli.



(*P. roqueforti* Thom) as characteristic of the French blue-mold cheeses. Our own investigation of this group of cheeses agrees with that of Sopp and Biourge in indicating that the various strains of the *P. roqueforti* series (Biourge's *stellata*) are practically always dominant in the interior of such cheeses.

The first species mentioned in Bainier's paper (already cited) was *P. vesiculosum* which remains unrecognized by others since 1907 and was unrecognizable in our own culture work until we examined (1926) what appeared to be a pathological culture of *P. roqueforti* (but undoubtedly belonging with *P. roqueforti*) and found a whole range of vesicular structures mixed with normal penicilli which clearly satisfied Bainier's description. Close rereading of Bainier's paper fails to disclose any suggestion that he recognized any mold of this group as an agent of cheese ripening. Nevertheless, one is almost compelled to believe that the Roquefort mold was included and to attribute his failure to write a recognizable description to his use of licorice sticks as the substratum for the colonies upon which he based his diagnoses. Of the other species described only one could satisfy the general requirement fairly well, *P. asperulum*, but other forms not connected with the cheese industry have often seemed to suit Bainier's diagnosis of *P. asperulum* better than *P. roqueforti*.

Even in the cheese industry considerable variation in the appearance of the molds used is evident in comparative culture (Golding). Members of this same series of races or species are found under many other conditions.

When numerous cultures from varied sources are examined the individuality of a particular strain appears less striking as variation after variation bridges the gap between organisms at first apparently separate. The names of the species known to belong in the series have been brought together with the characteristics ascribed to them by the describers. Forms like *P. suavolens* Biourge are fairly definitely separable from *P. roqueforti*, but the identification of newly found cultures as one of these species from the descriptions published by Sopp, Weideman, Thom, and Biourge is scarcely probable, although these descriptions may be illuminating as emphasizing the range of variations found and in a general way characterizing more or less definite sections of this great series of related organisms.

Biourge attributes the description of the type of this group to Dierckx and says that *P. atro-viride* Dierckx was intended to include all the fungi connected with the cheeses Roquefort, Gorgonzola, etc. He adds that

if a collective name is to apply to the whole series of closely related strains it must be *P. atroviride*, otherwise the whole series of names he proposes must be used for the strains to which they apply.

In general he believes that there are at least three and better five species in this group. These appear to be *P. roqueforti*, *P. gorgonzola* (= *P. weidemannii*), *P. suavolens*, *P. stilton*, and ? *P. virescens* Sopp ? a and b. Since no one was able to decide from his description what organisms Dierckx intended by his *P. atro-viride*, and since the cheese molds were adequately described and became well known in the period between Dierckx (1901) and Biourge (1923), Dierckx species name may justly be dropped.

160. *P. roqueforti* Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 82, pp. 35-36, fig. 2. 1906. See also idem Bul. 118, p. 34, fig. 4. 1910. See fig. 38.

Probable synonyms: *P. aromaticum* Sopp, *P. vesiculosum* Bainier. *P. glaucum* in the French cheese literature (see Duclaux, p. 694. Mazé, Roger).

Emended diagnosis: Colonies on Czapek's solution agar velvety with surface fairly smooth or plane, spreading broadly, with margin broad, white, thin, cobwebby or veil-like, with hyphae radiating partly on the surface partly just below the surface of the substratum, and green conidial areas following the hyphae in unevenly radiating lines (basis of Biourge's section name *Stellata*), forming a mass 100 to 300 $\mu$  deep, at margin white then bluish green and then quickly dull green; reverse in shades of green or bluish green to almost black, varying with conditions of culture; conidiophores mostly short about 100 $\mu$ , or, less frequently, up to 200 $\mu$  long, by 4 to 6 $\mu$  in diameter, ascending from aerial loops or submerged sections of vegetative hyphae, often branched with walls roughly granular and pitted (occasionally smooth); penicilli variously simple monoverticillate masses, verticils of metulae and sterigmata, compact branching systems, or with one or more long appressed or diverging branches with asperulate walls, sometimes giving a cymose appearance; metulae about 12-14 by 4 $\mu$ , with walls more or less evidently pitted or even asperulate; sterigmata up to 12 by 2 to 3 $\mu$ ; conidia globose or subglobose 4 to 4.5 $\mu$  less commonly 5 $\mu$  or larger (see Biourge's figures), in long chains forming loose to fairly compact columns.

Biourge in his monograph (La Cellule 33: fasc. 1. pp. 202-204; col. pl. V, cart. 155; pl. VIII, fig. 44. 1923) described his colonies on wort gelatine as green (C.d.C. 338) and gives measurements and details as

follows: conidiophore about  $4\mu$  in diameter; penicillus about 50 to  $70\mu$  long, with all walls smooth, figured as main axis with its terminal verticil of metulae, and one branch of irregular length bearing an accessory conidial mass; branches up to 25 by  $3.5$  to  $4\mu$ , single or in pairs; metulae about 15 by  $2.5$  to  $3\mu$ , in twos or threes; sterigmata 8 to 14 by  $2$  to  $3\mu$ , in groups of 2 to 6; conidia oblong 3 to 7 by  $2.5$  to  $6\mu$ .

Biourge's culture no. 155 (our no. 4733.102) when grown on Czapek's solution agar varies somewhat from Biourge's description, the colonies were green, without bluish tinge (except on wort agar) about 100 to  $200\mu$  in depth, but with stalks about  $100\mu$  by  $5\mu$ , rough as also the branches and metulae; conidia were 4, 4.5, 5 and almost  $6\mu$  subglobose.

In extending Thom's description of 1906, the inadequacy of that description is recognized together with the pertinence of the criticisms of Westling, Weidemann and Biourge. Nevertheless, the student of these molds is best served by the incorporation of the corrections in the description of this organism which is widely used in the cheese industry, rather than in the recognition of another species name to cover the cheese mold and the relegation of this name to a species of minor importance. The original description was prepared before the multiplicity of strains, varieties or species in this series was appreciated. Similarly considering this multiplicity of intergrading forms, hundreds of which have been handled, we are constrained to doubt whether any worker with the resources of a culture laboratory can positively separate *P. roqueforti* Thom, *P. gorgonzola*, and *P. stilton* Biourge. Our own experience covers many years of association with the cheese industry (see bibliography Thom) during which molds of this series have been isolated many times and thousands of cheeses have been made with cultures prepared from our stock strain (no. 18) originally isolated from French Roquefort cheese.

Within the cheese industry, as indicated by Sopp, and later by Weidemann, Bainier, Biourge, Steuart, Golding, Scaramella, Arnaudi and others, many cultures when compared show minor variations from any single type description. Golding working with the English types of blue mold cheese isolated and sent us a series of strains varying in individual details of structure and reaction and showed that the biochemical differences among them were great enough to affect their practical usefulness. Similarly, workers in this department have found changes in the strain of *P. roqueforti* used for inoculation to affect the quality of their cheeses. Arnaudi has made a similar study of molds from Gorgonzola cheese.

The following species with the general characters of *P. roqueforti* have been described. Except the species of Biourge which we have cultivated, such species as *P. virescens* Sopp, *P. atroviridum* Sopp, *P. griseobrunneum* Sopp, and *P. vesiculosum* Bainier have been placed here from analysis of the descriptions given. Comparison of these descriptions shows a considerable range in colors, depth of colony, measurement of elements, and in biochemical reactions. When large numbers of cultures of members of this group are compared, practically all of the data given in these descriptions are encountered in actual growing colonies, but very few of these descriptions are complete enough to insure a positive identification with some strain in culture.

161. *P. stilton* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 206-207; Col. Pl. V, Cart. 151; Pl. VII, fig. 42. 1923.

Colonies on wort gelatine, thin at first bluish green (glaucous), then grayish olive green, finally brown, coremia none; reverse pale greenish to orange, with areas of green and dark olive; odor *mildly cheesy*; conidiophore about 3 to 3.5 $\mu$  in diameter; penicillus mostly 30 to 40 $\mu$  long, with all walls smooth; branches 15 by 2 to 3 $\mu$  in pairs; metulae 8 to 15 by 2 to 3 $\mu$ ; sterigmata 6 to 11 by 2.5 to 3 $\mu$  in verticils of 2 to 6; conidia at first 2 by 1.5, soon 3 by 2, 3.5 by 2.7, finally globose 5 to 6; or 7.5 by 6 $\mu$ .

Biourge no. 151 (our no. 4733.116) was separated from *P. roqueforti* most readily by culture on potato upon which it grows poorly even less well than *P. gorgonzola*.

Upon Czapek's solution agar 4733.116 produces colonies with the general marks of the *P. roqueforti* series, but with more aerial hyphae, a tendency to floccosity, giving a very broad fibrous marginal area tardily developing pale gray green color with the ripening conidia; reverse colorless; hyphae coarse 6 to 9 $\mu$  in diameter; conidiophores rough with pitting fine but evident; conidia 4 to 6 $\mu$ , with the suggestion that the larger ones are swelling toward germination.

A culture (no. 4725.796) from C. G. Hansford in Jamaica satisfies this description as does no. 4825e from Pecorino cheese imported from Italy.

162. *P. atro-viride* Dierckx, Soc. Scientifique Bruxelles 25: 87. 1901.  
Synonym for *P. roqueforti* Thom.

*Dierckx's* brief characterization may be translated: Conidia 3 to 4 $\mu$ . Sterigmata 2 to 3 [presumably in the verticil]; penicillus 70 $\mu$  in length more or less; appearance powdery; conidia pure blue, blue green with

brownish shade, then violaceous brown; reverse clear green, then diffusely dark greenish brown; a typical form.

This description was hopelessly inadequate. With the aid of Dierckx's notes however Biourge (Monogr., p. 199) decided that Dierckx's description included in one series all of the forms related to the Roquefort cheese mold, hence decided that these forms must either be "lumped" under *P. atro-viride* or divided into 5 species as in Biourge's Subsection Stellata. Saccardo in the Sylloge, vol. 16, p. 1030, 1902, merely lists the species of Dierckx as briefly characterized with the hope that his complete publication would make possible the identification of his forms. Since the material has never been published and Biourge delayed his own publication until 1923, it seems best to reduce *P. atro-viride* Dierckx to the list of synonymy for this group of related forms.

163. *P. aromaticum* I (Roquefort) Sopp. Monogr. pp. 155-156, Taf. XVII, figs. 118, 119, Taf. XXII, figs. 7, 8. 1912. See also Centralb. f. Bakt., 1896, and Über Käsevergärung, p. 68.

Colonies upon meat-peptone-sugar-gelatine, clear green with fine even surface growth (velvety CT) and very fine hyphae (?—C. T.), spreading over the substratum; odor sweetish, attractive but quickly dulled; taste typical of Gammelost; temperature limits—15° to 35°C., even to 40°C.; liquefying wort gelatine in 11 days; grows well under partly anaerobic conditions with the production of green spores; casein fully dissolved with the odors of Roquefort and Gammelost.

Sopp's discussion of this organism is not sufficient to insure identification, although our general knowledge of the flora of these cheeses indicates a member of the *P. roqueforti* series.

163a. *P. aromaticum casei* Johan-Olsen [Sopp], in Centralb. f. Bakt. etc. 2 abt. 4 (no. 5) pp. 161-169, 1898; cited by Saccardo, Syll. 22: 1279 and attributed to "Sopp."

In this paper, the author discusses his cheese mold under this name but without describing it or definitely connecting it with a described form although it was probably one of his numbered strains. The name should be dropped. Similarly several other strains of Sopp's series of cheese molds are referred to as *P. aromaticum* with numerals, or modifying terms, such as "I," "II," "III," "Roquefort," "Gammelost." None of them can be identified any more closely than to the *P. roqueforti* series.

164. *P. roqueforti* var. *megalospora* (mss. species of Dierckx), Biourge Monogr., p. 203, 1923.

"Dierckx appelait dans son manuscrit var. *megalospora* le type du Roquefort."

Biourge's examination of Dierckx's old cultures gives conidia in the middle of chains  $5.5$  by  $4.5\mu$ , and the oldest ones  $7.2$  by  $6.5$  to  $7\mu$ .

We have seen such measurements in Golding's strain no. 40 sent to us about 1925.

165. *P. roqueforti* Thom var. *Weidemannii* Westling. Arkiv. for Botanik 11, pp. 52, 71-73, fig. 6 and 49, 1911; cited by Biourge Monogr. p. 204 as *P. weidemannii* Westling.

Variety differs from type in reverse of colonies green or dark green, not uncolored. Westling quotes Weidemann and cites his own experience that the reverse of the colony as found in their cultures from cheese is always green, whereas Thom described the reverse of *P. roqueforti* as uncolored. Weidemann and Westling are unquestionably correct in their observation of the green color in the reverse of most strains of *P. roqueforti* upon most media, but the observation should be made an emendation of Thom's description rather than the basis of a new variety.

166. *P. weidemannii* Westling. Biourge Monogr., p. 204. 1923.

Westling (p. 71) did not give *P. weidemannii* but *P. roqueforti* Thom var. *weidemannii*.

168. *P. biourgei* Arnaudi. Boll. Ist. Sieroterapico Milanese, vol. 6, fasc. I, p. 25-27, Pl. 1-2, 1927; see also Centralb. f. Bakt., 2 Abt. 73 (15/23): 321. 1928.

Not *P. biourgei* Dierckx Soc. Scientifique Bruxelles 25: 88. 1901; probably *P. roqueforti* q. v.

In culture on eighteen different media, surface blue green to dark chocolate with age; reverse yellow rose, yellow, grayish orange to dark gray yellow; vegetative hyphae hyaline, filamentous, septate,  $6$  to  $6.5\mu$  in diam.; fruiting in three days on acid glucose agar; stalk hyaline, septate, branching three times with branches running nearly parallel and close together; metulae first set  $11.25$  to  $13.75$  by  $5$  to  $5.5\mu$ , each with two or three branches,  $8.75$  to  $10$  by  $2.5$  to  $2.75\mu$ ; sterigmata, two to three on each of second set of metulae, somewhat prolonged or fusiform,  $11.5$  to  $13$  by  $2$  to  $2.5\mu$ ; penicillus  $30$  to  $37.5\mu$  in length; conidia globose, rarely suboval, smooth, glaucous,  $3$  to  $5\mu$  in diam.

On acidulated glucose broth, liquid clear yellow, reverse yellow, surface chocolate when old, border white spreading upon flask. On potato with Roux solution, edge yellow and red when young, later red disappears and whole surface fruits giving a green color, but yellow where in contact with glass. On milk yellow color but not diffused thruout the milk, digestion total.

Arnaudi marks his *P. biourgei* again as "n.sp." in his German article in the Centralblatt and gives an inadequate diagnosis in Latin which is freely translated as follows: Sterile hyphae hyaline septate 6 to 6.5 $\mu$  in diameter; conidiophores hyaline, with branches in threes, septate, parallel, almost decurrent 11.25 to 13.75, 5 to 5.5 $\mu$ , penicillus 30 to 37.5 $\mu$  long; metulae in twos or threes 8.75 to 10, 2.5 to 2.75; conidia globose, rarely suboval, smooth, glaucous 3 to 5 $\mu$ .

Although efforts have been made to secure Arnaudi's strains and they were promised to us at one time, we have not received them. We are not convinced that the species of Arnaudi is sufficiently distinct from *P. roqueforti* to warrant separation. An abstract of Arnaudi's second paper follows.

Arnaudi, Carlo. Über die Penicillien des Gorgonzolakäses. Centralbl. Bakteriöl., Parasitenk. Abt. 2. 73(15/23): 321-330, 2 fig. 1928. —Arnaudi reviewing his previous work, gives a Latin diagnosis for *Penicillium Biourgei* as new and refers the other fourteen races of molds found in the cheese to *P. Weidmanni* Westling var. *fuscum* already discussed. In agglutination reactions, six strains of *P. Weidmanni*, *P. Biourgei*, *P. "tossico"* of Carbone and Cazzomalli, and *P. "Giallo"* of Ventruelli were found specific for the four separate species tested, and only slightly less so for the races of *P. Weidmanni*. A series of biochemical tests of the six races of *P. Weidmanni* and *P. Biourgei* included optima and tolerances toward sodium chloride, lactic acid plus optimum sodium chloride and sodium chloride plus optimum lactic acid, the rate of digestion of milk and the colors produced in the substratum after one month. Each race tested produced a different shade of color, the optimum for lactic acidity varied from 2 to 6 per cent. While the results point toward practical application to the Gorgonzola cheese industry further work will be required.

169. *P. weidmanni* Westling, var. *fuscum* Arnaudi. Boll. Ist. Sieroterapico Milanese, vol. 6, fasc. I, p. 18-25, 26-27. 1927.  
Varietal name assigned on page 27.

Arnaudi states that Biourge assigns to Grossbüsch's culture of a strain

of mold from Gorgonzola cheese the name *P. gorgonzola weidmanni* or *P. weidmanni* Westling.

Arnaudi describes these strains as varying from others by having no rosy color, and browning potato entirely.

170. *P. virescens* Sopp. Monogr., pp. 157-159, Taf. XVII, fig. 121; Taf. XXII, fig. 4, 5. 1912. See fig. 39.

Colonies on meat-peptone-sugar-gelatine, at first green, later blue green without yellowish tinges, with brownish lines, velvety spreading evenly over the substratum, without liquefying the gelatine; mycelium thin, easily torn, somewhat veil-like; hyphae coarse, vacuolate; reverse dark almost black-green; odor suggestive of burnt bread, on milk becoming "cowy," on wort aromatic; conidiophore figured as short, coarse, with penicillus consisting of the stalk, appressed branches, metulae, sterigmata and conidial chains forming a compact mass only slightly splitting toward the ends of the conidial mass; conidia globose, smooth, 6 to 8 $\mu$  (p. 157) and 6 to 7 $\mu$  in table, p. (134 and p. 158); perithecia not found.

Species found in earth. Cultures grew best at 30°, with a minimum at 5° and maximum higher than the related forms, 38 to 40°C., and grew well in all common media tested. Conidia remained viable more than three years. The measurements and appearances given are more or less satisfied by a vigorously growing member of the *P. roqueforti* series isolated from Pecorino cheese (our no. 4825s).

The name is untenable because even though we were to find Sopp's species, the name had been already used by Bainier in 1907 for a different species.

171. *P. suavolens* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 200-202; Col. Pl. V, Cart. 7; Pl. VIII, fig. 4. 1923.

Colonies on wort gelatine, forming a thin layer, pale bluish green, with broad margin arachnoid (ajourée), coremia none; reverse yellowish; odor pleasantly sweetish; gelatine liquefied; conidiophore 3 to 4.5 $\mu$  in diameter (figured as smooth) penicillus usually about 40 $\mu$ , occasionally 70 to 100 $\mu$  long, with all walls smooth (compare our cultural description) figured as main axis, terminal verticil of metulae varying greatly in length and occasionally septate and producing secondary branches 12 to 15 or up to 25 by 2.5 $\mu$ , in twos or threes; metulae 8 to 12 by 2.2 to 3 $\mu$  in verticils of 2 to 4; sterigmata 8.5 to 12 by 3 to 3.5 $\mu$ ; conidia subglobose to globose variously from about 4 $\mu$  up to 6.5 by 5 $\mu$ .



Biourge no. 7 (our no. 4733.118) as received and grown by us is a characteristic form but varies enough from Biourge's description to require a full description as it appears in our culture! Our notes follow:

Colonies of 4733.118 on Czapek's solution agar, azonate, velvety, broadly and rapidly spreading more or less buckled or wrinkled in center, forming an aerial mass 100 to 300 $\mu$  in depth with margin broad and arachnoid, conidial areas pale gray green becoming mouse gray in age, with scanty overgrowth of cottony tufts; reverse colorless at first then pale yellow and brownish in age; odor faint or doubtful; conidiophores commonly 100 $\mu$  or less, occasionally as trailing hyphae much longer, 3 to 5 $\mu$  in diameter with coarsely granular walls, arising either above or below the surface of the substratum; penicillus either a terminal verticil of metulae, rarely single and citromyces-like or a main axis with 1 or 2 branches bearing verticils of metulae, and sterigmata about 10 by 2.5 $\mu$  with chains of conidia masses into columns often up to 200 $\mu$  or more in length; conidia subglobose mostly about 4 $\mu$  in long axis ranging from 3 to 5 $\mu$  and germinating at about 5 $\mu$  by several tubes not arising at the poles of the conidia.

Cultures approximating Biourge's species were received from Miss Dale in 1912 (no. 2692), Miss Rose in Oregon (no. 4390.27B<sub>2</sub>), from Hood River on fruit boxes (no. 4644), from Bristol, England on tobacco no. 4742 P. 9, and from the pathologists studying the rots of sugar beets in storage in Colorado (no. 4975.109,—.123 and .136).

172. *P. gorgonzola* Weidemann. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 204–206; Col. Pl. V, Cart. 154; Pl. VIII, fig. 43. 1923.

Synonym: *P. roqueforti* var. *weidmanni* Westling, p. 71.

Colonies in wort gelatine, thin, broadly spreading, at first blue green (C.d.C. 397), then dark green, finally brown, coremia none; reverse at first pale yellow, then reddish yellow, with green areas becoming almost black in age; odor weak, sweetish; conidiophores 3 to 5 $\mu$  in diameter; penicillus 50 to 100 $\mu$  long, with all walls smooth, figured as a coarse main axis with terminal verticil of metulae and one or more branches at the second node producing accessory conidial masses; branches 15 to 20 by 3 to 5 $\mu$ , in twos or threes or again branched; metulae 12 to 14 by 3 to 4.2 $\mu$  in twos or threes; sterigmata 7.5 to 12 by 3 to 3.5 $\mu$ , in verticils of 3 to 5; conidia at first oblong 3.5 to 4.8 by 2.5 to 4 $\mu$ , at length nearly globose 6.5 by 5.5 to 5.8 $\mu$ , figured as varying enormously in size.

Biourge's no. 154 (our no. 4733.65), was received as *P. gorgonzola* from M. Grossbusch at Ettelbruck. He notes that this form grew poorly on potato, on which *P. roqueforti* grew well.

On Czapek's solution agar no. 4733.65 produced broadly spreading densely velvety colonies uneven at the arachnoid margin, about 200 to 300 $\mu$  in depth, with conidial areas at the margin pale bluish, then blue green, with reverse uncolored, with stalks short about 4 $\mu$  in diameter, pitted (rough); and conidia 4 to 5.5 $\mu$  subglobose.

If the name is to be used for blue-green members of the series, many cultures will be found which will in general satisfy this description. Our no. 3515B from rancid butter reproduces the details of Biourges diagnosis.

173. *P. atro-viridum* Sopp. Monogr., pp. 149-150, Taf. XVI, fig. 114; Taf. XXIII (XXII corrected), fig. 12. 1912.

Colonies blue-black, in age black-gray or black-brown, with abundant conidial areas spreading gradually over the substratum without marked wrinkles; mycelium is at first white, reverse in age black; odor strong; conidiophores coarse, fairly short, partly branched, articulate (gegliedert) each cell of the stalk increasing in diameter toward its apex, and the stalk somewhat vesiculose at the apex, penicillus figured as consisting of a branch or verticil of branches, then metulae, short somewhat swollen, and sterigmata with conidial chains adherent into columns; sterigmata large, flaskshaped; conidia globose, uneven in size, 4 to 6 $\mu$  diam., angular, blue-black, in short chains and tending to mass into balls as in *Gliocladium* (? column formation—C. T.); perithecia not found.

Species obtained from cultures from the atmosphere; colonies grew well in gelatine and agar media, in milk surpassingly richly, coloring the fat more or less yellow and coagulating it to a flocculent, bitter curd; good growth also in broth, in wort, on potato, on bread; conidia remained viable more than 3 years in the laboratory.

No culture we have seen satisfies Sopp's specification of *angular* conidia.

174. *P. roquefort* Sopp. Monogr., pp. 156-157, Taf. XVII, figs. 118-119; Taf. XXII, fig. 7, 8. 1912.

Colonies on meat-peptone-sugar-gelatine, with veil-like almost Mucor-like appearance of the growing colony, clear bluish green, later chocolate brown; reverse at first colorless, later reddish or greenish at times; odor at first sweetish somewhat insipid, later cheesy; conidio-

phore coarse, figured as smooth, with penicillus figured as consisting of 1 divergent branch, and then metulae and sterigmata; conidia reported in variety *a* as rarely over  $5\mu$  diameter, in variety *b* up to  $6\mu$ ; perithecia not found.

Species characteristic of the firm cheeses which show green mold in their interior channels or cavities. Cultures grew best at  $20^{\circ}$ , with minimum of  $5^{\circ}$  and a maximum at  $35^{\circ}\text{C}.$ , and grew well on all the common media reported.

Sopp cites the same illustrations for *P. aromaticum* and *P. roquefort*. Without referring to Thom his description shows a familiarity with Thom's diagnosis of *P. roqueforti*.

175. *P. aromaticum* "gammelost" Sopp. Monogr., pp. 159-161, Taf. XVII, fig. 123; Taf. XXII, fig. 10. 1912.

Synonym: *P. aromaticum* II.

Colonies on meat-peptone-sugar-gelatine, clear yellowish blue-green then yellowish green in age, finally gray green to brownish, with wrinkled mycelium at first veil-like then in reverse white, then greenish, or reddish in wort cultures, often splitting in growth to leave sterile areas; conidiophores moderately coarse and fairly long, figured as smooth, with penicillus figured as consisting of main stalk and fairly long slender appressed branch or branches, close verticils of fairly long metulae and sterigmata with chains of conidia tending to remain in columns; conidia globose, smooth  $4$  to  $7\mu$ ; perithecia not found.

Species described as characteristic of the sour-curd type of cheese—Gammelost—in Norway and represented by several varieties with different biochemical activities, hence greatly different influence upon the cheese. Cultures grew best at  $20^{\circ}$ , with minimum at  $3^{\circ}$  and maximum at  $33^{\circ}\text{C}.$ , and grew well upon all common media tested. Conidia remained viable more than five years. Sopp separates this form from the *P. roquefort* types by its predominantly yellow green color, slight color in reverse, lower temperature maximum, and different action upon cheese.

176. *P. griseo-bruneum* Sopp. Monogr., pp. 153-155, Taf. XVII, fig. 117; Taf. XXII, fig. 6. 1912.

Colonies on meat-peptone-sugar-gelatine, bluish gray becoming gray brown in age (upon potato deep green with Valerian odor); indicated by figures as velvety; mycelium without wrinkles, close textured rather thin; with coarse hyphae; reverse greenish gray brown, in milk becoming

dark blue-gray; hyphae coarse; odor on most media slight or indefinite; conidiophores coarse, long, granular, with penicillus consisting of long, more or less diverging branches, metulae and sterigmata, with walls granular or rough; sterigmata described and figured as long and said occasionally to be flaskshaped; conidia large, globose, smooth, yellow brown, 7 to 8 $\mu$  in diameter in long chains.

Species found parasitic upon a *Stereum*. Cultures showed a temperature optimum of 20°, a minimum of 8° and a maximum of 33°C. Good growth was obtained upon all common media.

The distinguishing character was conidia smooth, 7 to 8 $\mu$  in diameter yellow brown.

177. *P. vesiculosum* Bainier. Bul. Soc. Mycol. France 23: 10-12; Pl. II, fig. 1-8. 1907. See fig. 39.

Colonies forming floccose tufts (on licorice sticks ?) pale green to dark green. Vegetative mycelium consisting of septate, branching, anastomosing hyphae at first cylindrical, but as fruiting begins, enlarging irregularly to form large vesicles (vacuoles ?), globose or variously elliptical, sometimes isolated, sometimes crowded together, until whole hyphae appear to be filled with big vacuoles crowded end to end producing the appearance of false septa between them; aerial hyphae and conidiophores also show vesicular enlargements and abundant vacuoles, which sometimes appear also in the elements of the penicillus; conidiophores figured as smooth; penicillus described and figured as composed of few but irregular branches, with metulae and sterigmata few in the verticil; metulae 3 to 4 in the verticil; sterigmata about 7 $\mu$  long, in verticils of 3, 4 or 5; conidia about 3.7 $\mu$  in diameter.

Species reported from a potato. Study of Bainier's description in connection with the paragraphs preceding this description which refer to the powdered bread prepared for Roquefort cheese-making, caused us to compare Bainier's figures with preparations made from variant members of the *P. roqueforti* (Stellata of Biourge) series. One of these cultures reproduced Bainier's figures so fully as to convince us that Bainier had under observation some member of the series, but our culture showed appearances which we believe to be pathological. We conclude, therefore, that *P. vesiculosum* as a normal species probably does not exist.

*Sub-section 5. Velutina-asperula.* Species differing from the *Stellata* in the lack of the veil-like or cobwebby (arachnoid) margin; broadly

spreading, azonate or at most hemizonate; walls pitted or rough; conidia subglobose.

Strains are occasionally encountered which differ from the Radiata in having conidiophores with definitely asperulate walls and differ from the Stellata in their regular outline and the absence of the broad cobwebby (arachnoid) marginal area in the growing colony. Although we have never been able to be certain of Bainier's *P. asperulum* it falls by description in this place and has been used as a series name for these strains in spite of a doubt whether Bainier's original culture was really separate from the Stellata or not.

180. *P. asperulum* Bainier. Bul. Soc. Mycol. France 23: 17, Pl. IV, fig. 13-18. 1907. See Westling, Arkiv f. Bot. 2: 140. 1911.

Colonies on licorice sticks at first pale blue, then blue green, in age dark sordid green, darker than *P. puberulum*, which is nearly related; hyphae vacuolate, averaging  $5.6\mu$  in diameter; conidiophores markedly sinuous or undulating, showing abundant crystalline granulations on the walls (? pitted), often bearing divergent (perpendicular) branches distant from the primary penicillus; penicillus figured as a main branch and partial verticil of short metulae, then sterigmata and conidial chains, parallel or in adherent masses; sterigmata and metulae not described but figured as few in the verticil; conidia globose, about  $4.2\mu$  in diameter, swelling very greatly in germinating and putting out large tubes.

Species reported as related to *P. puberulum* but darker in color; original source not reported. Westling does not appear to have identified a culture. Biourge does not even report the species. No other worker has proposed to use the name for a well-identified organism.

## CHAPTER XV

### THE BREVI-COMPACTA

*Asymmetrica* Section 2. *P. brevi-compactum* series (Hemizonata Biourge). Colonies partly floccose, partly lanose, partly velutinous; penicillus terminal and biverticillate with or without a second branch and verticil showing a characteristically short compact base with diverging sterigmata and conidial chains (fig. 40).

Dierckx's name *P. brevi-compactum* as discussed and figured in Biourge's monograph is descriptive of the penicillus in a series of forms represented by *P. paxilli* Bainier, *P. stoloniferum* Thom, *P. tabascens* Westling and probably *P. crassum* Sopp. Unfortunately Dierckx's

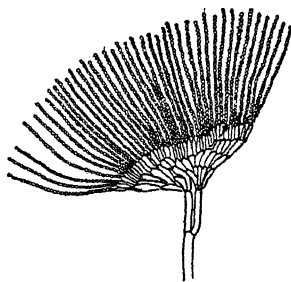


FIG. 40. The Brevi-compactum type of penicillus: Note the compact base with chains of conidia parallel to divergent; an extreme figure showing a third or fourth verticil in some parts of the penicillus but such figures occur in the series.

description was too meagre to identify the species. With the cultures of Bainier, Westling, and Biourge to compare with Thom's type, and with other accumulated strains having this morphology, certain characters seem consistently enough present to warrant holding these organisms in a section separate from the Biverticillata-symmetrica to which they bear some resemblances.

Some other observations of a more or less general application may be useful. The colonies upon Czapek's solution agar are commonly azonate, or in some tardily show zonation especially toward the margin (hemizonate of Biourge). They vary from floccose to almost velvety

mostly with some aerial felt of hyphae in the central area at best. Velvety and thinner margins are characteristic of part of the species.

Stolons or marginal aerial hyphae extending beyond the submerged mycelium and returning to the substratum were figured by Thom for *P. stoloniferum* (fig. 41) and have been seen in many strains of this series when grown upon very wet media in humid containers. All of the species studied carefully have liquefied gelatin so rapidly that the colonies quickly floated in pools.

Conidiophores arise either from submerged or from aerial hyphae; they are short in some, very long in other strains or short and long conidiophores are both found in some strains.

The penicilli may consist of terminal verticils of metulae in short very compact groups with crowded sterigmata compacted at the base and more or less diverging at the apex, and with chains of conidia parallel, more or less diverging and in age becoming tangled masses, or part of them may have in addition to the terminal verticil one to three branches at the next node bearing metulae closely compacted to the primary terminal verticil of metulae and penicilli partially or more or less completely repeating its structure or with one diverging branch bearing a secondary verticil (or penicillus) repeating the structure of the primary verticil.

Conidia in the group are globose or subglobose and small—rarely up to  $4\mu$  in long axis.

*P. stoloniferum* Thom and *P. parilli* Bainier were both described from rotting mushrooms. The stolon-like margin was observed by Thom on the rotten area of the original mushroom. Many cultures have since been seen from soil and decaying substances. The structure of the penicillus appears however as a much more stable character than stolon production which was originally thought to be significant. Stolons do, however, appear in most of these species as grown in moister situations or upon softening media.

As included here the penicillus with short compact base, and the sterigmata with short abrupt tubes (instead of the lanceolate tubes of the *Biverticillata symmetrica*) separate these species from the divaricate series on the one side and the *Biverticillata symmetrica* on the other. Cultures showing this type of penicillus with reactions and habits allying them with *P. stoloniferum* or to the *P. brevi-compactum* group to use Dierckx's name, have been received from many sources only a few of which may be listed here to show the wide distribution of the sources of this material.

4662.2	from Missouri Botanical Garden
4178	" Ludwig
6 (cultures)	" Miss Rose, Corvallis, Oregon
2701	" soil England, by Miss Dale
4737.3	" Iowa—Abbott
4759	" England, 5 strains, Mrs. Kidd
4601	" Maine on <i>Vaccinium</i> , N. E. Stevens
4315.B10	" Lincoln, Nebraska
4406.49	" Syracuse, New York, on paper stock
4640.451	" France <i>P. paxilli</i> Bainier
4235.18	" China
4707.7551	" Brazil, Fonseca
4356	" Core rots of apples
4200.4	" Imported filberts

Turesson working in Sweden found some members of this series pathogenic to honey bees and discussed the symptoms produced in experiments. When fed in fairly large quantities all of his experimental bees were dead in eight days. His further experiments tended to indicate that a phenolic substance was produced in the medium in sufficient quantity to produce the toxic effect.

#### KEY TO SPECIES OF THE *P. brevi-compactum* SERIES

Colonies when young stolon forming (mostly); rapid liquefiers of neutral gelatin media; with the short abruptly compact base and diverging or parallel chains of conidia (not in columns).

- I. Conidia globose or subglobose.....II.
- I. Conidia elliptical.....X.
- II. Colonies forming fairly deep mycelial masses with long conidiophores.....III.
- II. Colonies approaching velvety; conidiophores short.....IV.
- IIIa. Colonies in yellow and green shades. Conidia 2.8 to 3.4 $\mu$ , smooth or very faintly spinulose in age; conidiophores up to 300 $\mu$  long.....*P. stoloniferum* Thom, 185.
- IIIb. Colonies dark blue green. Conidia about 2.8 $\mu$ ; conidiophores up to 1,000 $\mu$  long.....*P. paxilli* Bainier, 186.
- IIIc. Colonies grayish blue to sordid green. Conidia about 2.8 $\mu$ ; conidiophores long.....*P. erectum* Bainier, 187.
- IIId. Conidia about 3 $\mu$ ; center deep, up to 1 mm. or more; margin narrow velvety.....*P. brevi-compactum* Dierckx-Biourge, 188.



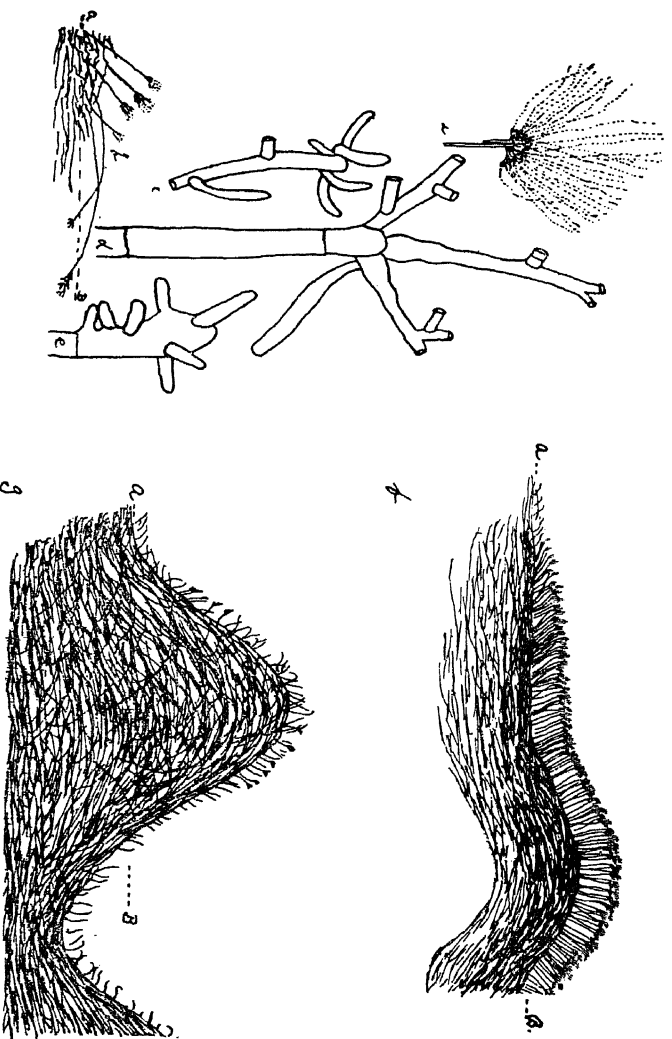
- IV. Colonies predominantly velvety; conidiophores short.....V.
- Va. Colonies blue green to coffee brown; conidia 2.5 to 3 $\mu$ .....*P. crassum* Sopp, 190.
- Vb. Colonies narrowly restricted in growth, about 200 deep, velvety, buckled and wrinkled; conidia 2 to 3 $\mu$  diameter faintly spinulose.....*P. Biourgeianum* Zal., 189.
- Vc. Colonies moderately broadly growing, velvety, showing zones 0.5 to 1 mm. wide toward the margin; dull green or olive; conidia about 3 $\mu$ ....*P. Hagemi* Zal., 191.
- Vd. Colonies thin, velvety, radiately wrinkled, yellowish green; conidia 2.5 to 3.5 $\mu$  diameter.....*P. Szaferi* Zal., 192.
- X. Conidia elliptical.....XI.
- XI. Colonies fairly deep, tending to floccosity.....XII.
- XI. Colonies velvety or nearly so in effect.....XIII.
- XIIa. Colonies bluish green at first then green and at last yellow green; conidiophores up to 1 mm. long, smooth; conidia 2.5 to 3 $\mu$  in long axis smooth or delicately roughened.....*P. tabascens* Westling, 193.
- XIIb. Colonies bluish green, faintly zonate in age; stoloniferous; about 300 to 350 $\mu$  deep; conidia 3 by 2 to 2.5 $\mu$ .....*P. aurantio-griseum* var. *Poznanensis* Zal., 194.
- XIIIa. Colonies bluish gray green to slate olive in age; 200 to 300 $\mu$  deep; conidia 3 to 4 by 2.5 to 3 $\mu$ .....*P. griseo-brunneum* Dierckx, 195.
- XIIIb. Colonies yellowish green to purple brown in age; conidia 3.5 $\mu$  in long axis, more or less elliptical...*P. patris-mei* Zal., 196.
- XIIIc. Colonies gray green to bluish green to brown in age up to 500 to 600 $\mu$  deep; conidiophores up to 600 $\mu$  long faintly pitted; conidia 3 to 3.5 $\mu$  by 2 to 2.5...*P. Bialowiezense* Zal., 197.

*Conidia globose. Mycelium forming deep masses with long conidiophores.*

185. *Penicillium stoloniferum* Thom. U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118: p. 68-69, fig. 26. 1910. See fig. 41, 42.

Colonies in gelatin or potato agar, green or yellowish green, becoming gray-green or gray when old (remaining green in sugar media), floccose,

FIG. 41. *P. stoloniferum* Thom (from Thom 1910): *a*, diagram of typical penicillus; *b*, habit diagram showing "stolon" formation at margin; *c, d, e*, submerged tips of "stolons" as they resume vegetative growth in the substratum; *f*, *g*, diagrammatic radial sections, *f* from a colony almost velvety and plane; *g*, from a colony wrinkled and buckled as in the photograph, figure 42.



spreading more rapidly in young cultures by aerial stolons than by submerged hyphae (i.e., the submerged mycelium seems to arise from the aerial rather than vice versa); reverse of colony not colored or partly yellow; conidiophores arising as short branches ( $100\mu$  or less in length) from aerial hyphae, or arising separately  $300\mu$  or more in length especially at the margins of older colonies; penicilli 40–80 more rarely up to  $170\mu$  in length, composed of short appressed branches and numerous sterigmata densely crowded at the base bearing very loosely divergent chains of conidia; sometimes the lowest branch diverges so that the penicillus appears double; sterigmata 10 by  $3\mu$ ; conidia slightly elliptical or globose,  $2.8$ – $3.4\mu$ , smooth, yellowish green in mass, almost hyaline by transmitted light; colonies liquefy gelatin very rapidly, with a strong alkaline reaction to litmus.

Habitat: Decaying fungi, Boleti, Polypori; cultures from milk and ensilage. Collected repeatedly at Storrs, Conn.; once upon decaying *Boletus scaber* at the Jardin des Plantes in Paris, hence probably widely distributed.

186. *P. paxilli* Bainier, Bul. Soc. Mycol. France 23: 95–96; Pl. X, fig. 1–4. 1907.

Colonies on licorice sticks, dark bluish green, conidiophores typically long, stiff, septate, averaging 1 mm. in length by  $2.8\mu$  in diameter, with shorter individuals and simple conidial apparatus frequent, especially in young colonies; penicillus described as typically a terminal verticil of 4 to 8 metulae, symmetrically arranged and each bearing 3 to 6 sterigmata but in other cases, producing a single branch at the next lower septum, equalling or surpassing the main axis in length, also producing 1 lateral branch and each terminated by a verticil of 3 to 6 sterigmata; sterigmata about  $8.4\mu$  long; conidia smooth, green, globose, averaging about  $2.8\mu$  in diameter, swell slightly in germination and emit several tubes.

Bainier's strain was obtained from a moldy Paxillus. It is probably represented by culture no. 4640.451 which came to us in the Bainier collection labeled *P. patulum*, but certainly was not that species, while it does approach Bainier's description of *P. paxilli*. This culture belongs to the series described by Thom as *P. stoloniferum* which was originally isolated from rotting mushrooms at Storrs, Connecticut. In any case the description given by Bainier specifies the details of branching and the number of branches far too definitely to be reproduced regularly by actual mold growth. It is easy to find an individual

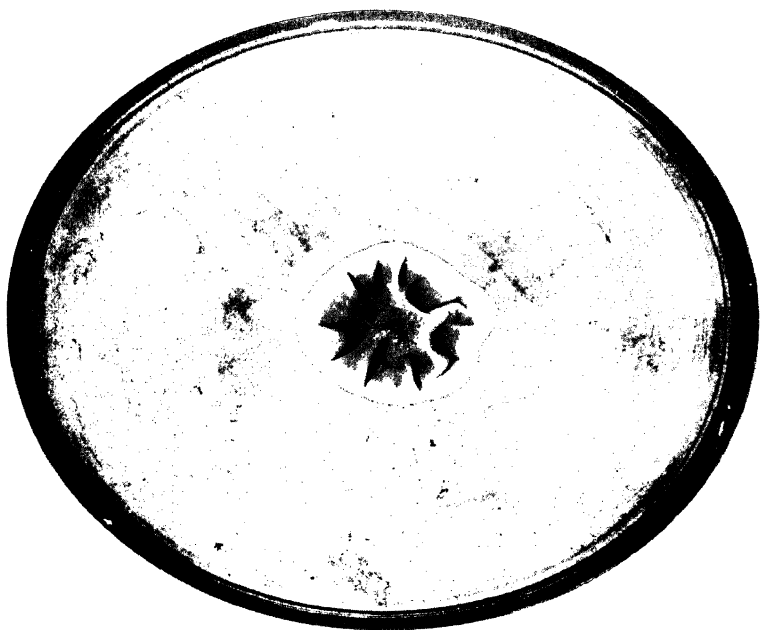


FIG. 42. *P. stoloniferum* Thom: Photograph of a wrinkled buckled and partly floccose colony.



penicillus to comply with the description, but the selection used by Bainier is more or less unfortunate as establishing a type.

187. *P. erectum* Bainier. Bul. Soc. Mycol. France 23: 13, Pl. III, figs. 1-16. 1907.

Colonies on licorice sticks produce floccose aerial mycelium pale ashy blue, then dark green; conidiophores long, unbranched, about  $5.6\mu$  in diameter, septate, arising as branches of aerial hyphae, penicillus with 1 or 2 series of branches in 1-sided verticils, then verticils of metulae and sterigmata; sterigmata up to  $19\mu$  long; conidia globose  $2.8\mu$  in diameter, swelling very greatly in germination and produce 1 to several tubes.

Species found on dead osier twigs in France as tufts of long rigid *Aspergillus*-like stalks terminated by penicillate spore masses, and Bainier notes that when the first penicillus has matured a new stalk branches out near the base of the old stalk to produce a second stalk a little shorter than the first, with secondary penicillus.

*P. erectum* is reputed by Bainier as common upon diverse substances. An occasional culture with the very long slender stalks described here, has been studied without reaching any satisfactory conclusion as to the definiteness of the species among the members of the group.

188. *P. brevi-compactum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88. 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 155-157; Col. Pl. II, Cart. 42, 1923; Plate III, fig. 16<sup>1</sup> is labeled for this species.

Synonyms: *P. paxilli* Bainier and *P. stoloniferum* Thom.

Colonies on wort gelatine, hemizonate (non-inflata?), with marginal zone 1 mm. broad submerged, then a narrow white zone changing to blue green, green and finally yellowish green in the conidial areas, coremial forms suggested in some media, for example, potato; reverse pale yellow; gelatine liquefied; conidiophores 4 to  $4.5\mu$  in diameter; penicillus 30 to  $40\mu$  long; primary branches about  $15\mu$  long in pairs or threes; metulae about  $13\mu$  long in verticils of 3 to 5; sterigmata 8 by  $2\mu$ , in verticils of 4 or 5; conidia globose  $3\mu$ .

<sup>1</sup> "Plate III, fig. 36" is referred to as an error in the text, p. 155, but the correction is not completed. Evidently "36" is a misprint for 16; the figure probably represents one of this group but in the face of Biourge's repudiation it can hardly be cited as part of the description.

Biourge's no. 42 (our no. 4733.21) upon Czapek's solution agar and in gelatine is clearly a member of the series falling under *P. parilli* Bainier and *P. stoloniferum* Thom. Dierckx's description in 1901 is utterly inadequate as a basis of identification hence should not be recognized although the name is beautifully descriptive.

The following notes from culture are significant: colonies on Czapek gray green to fairly deep green, velvety, loose, long stalked, upon a thin basal aerial felt up to 1 mm. in depth in center, with margin of conidial area velvety and a very narrow white margin; on gelatine yellow green quickly liquefying and producing the marginal stolons described by Thom: conidiophores 4 to  $6\mu$  in diameter, obscurely pitted, penicillus with short appressed primary branch, crowded and more or less diverging metulae and sterigmata with loosely parallel then tangled chains of conidia 3 to  $3.5\mu$  in diameter.

*Conidia globose. Colonies tending toward velvety; conidiophores short.*

189. *P. Biourgeianum* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 462, 463, 464; Taf. 45 and 48; Zaleski no. 1477.

Colonies on neutral Raulin with 10 per cent gelatine, about 30 to 35 mm. in diameter after twelve days, forming a symmetrical mass elevated 3 to 5 mm. above the substratum, with gelatine unchanged during the first twelve days but later entirely and quickly liquefied, surface growth velvety, azonate, with a broad marginal area white, then a pale blue green ring, C.d.C. 428B about a central area partly depressed, partly elevated, and in blue green shades such as 367, 372, or later green 342, 343; reverse in pale yellow to orange colors; conidiophores about 500 to 700 by 3 to  $4\mu$ , straight or slightly flexuous, simple or rarely branched, with apices usually inflated, with all walls smooth; branches 25 to 30 by 3 to  $4\mu$  rarely produced as one to three unequal members of a group at the apex of the conidiophore; metulae about 12 to 16 by  $2.5$  to  $3.5\mu$ , commonly 4 to 6, unequal in length in the verticil, with apices inflated to 4 to  $5\mu$ ; sterigmata about 9 to 10 by  $2.5$  to  $3\mu$ , commonly 8 to 12 in the verticil, straight or incurved with long tubes; conidia 2 to  $2.5$  or  $3\mu$ , smooth, globose with connectives distinctly seen in the chains.

Habitat: Species isolated from earth in mixed woods at a depth of 5 to 8 cm. in square 373 of the forest known as "Puszcza Bialowieska." Biourge assigned the culture sent him to the section with *P. chrysogenum*

Thom, and *P. brunneo-rubrum* Dierckx. Biourge is probably wrong. The description and figures given do not suggest a *Biverticillium* nevertheless Zaleski calls it "*Biverticillium* Dierckx subsec. 1 *Colliformiter-elevata*." Our notes follow: Type strain growing well about 20°C., very poorly at 30°C., or above; colonies upon Czapek's solution agar at 20°C., restricted in growth about 10 mm. in diameter in seven days, velvety with central area buckled or with cerebriform wrinkles, and a few deep radiate wrinkles thinning to a very narrow white margin about 1 mm. wide with contour more or less crenulate, velvety forming a mass not over 200 $\mu$  deep, transiently bluish green to green (about C.d.C. 343) in a narrow band at the edge of the conidial area passing to olive green, gray green shades and finally becoming fawn in about four weeks (Ridgway XL); reverse slowly developing yellowish green shades and later shades of orange; odor in old colonies strong, unpleasant; conidiophores 100 to 200 $\mu$  by 2 to 3 $\mu$ , ascending rather than erect, with apices enlarged or inflated; penicillus either a single verticil of metulae or with one branch 10 to 15 $\mu$  in length (usually shorter than the main axis) at the first septum producing metulae crowded at the base and diverging at tip, with long, loosely diverging or coiling chains of conidia breaking up in mounts; metulae up to 10 to 12 $\mu$  closely packed at base, diverging at apex; sterigmata about 10 by 2.5 $\mu$ ; conidia 2 to 3 $\mu$  in diameter, pale brownish, smooth during the growing period, faintly spinulose or granular in old colonies (reported as smooth by Zaleski).

Culture no. 5010.5 received from Baarn in July, 1928, is probably type.

190. *P. crassum* Sopp. Monogr., p. 147-148, Taf. XVI, fig. 111; Taf. XXII, fig. 15. 1912.

Colonies on meat-peptone-sugar gelatine blue green, then coffee brown, with heavy wrinkled or folded (buckled) mycelium; reverse gray to brownish; hyphae coarse; conidiophores moderately coarse, branched, comparatively short (figured as enlarging upward); penicillus with short branches each swollen at the apex and bearing a monoverticillate group of sterigmata (figured as first bearing a divergent branch then verticils of divergent rather short, heavy metulae and chains of conidia; sterigmata tending to be long and narrow, irregularly produced and irregularly shaped, and occasionally branched; conidia 2.5 to 3 $\mu$ ; perithecia not found; bodies suggesting sclerotia occasionally produced upon rice.



Species found upon a rotten apple with optimum temperature about 20°, minimum 3° and maximum 30°C. Colonies grew well on gelatine and agar, on milk producing yellowish mycelium and olive green, velvety conidial area, grew well also on potato, rice and bread; no coloring substances were produced; the odor varied with the substratum without being positively diagnostic. Conidia remained viable for more than three years.

191. *P. Hagemi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et. Nat. Ser. B, 1927, pp. 448, 449, 450; Taf. 39, Zaleski no. 579.

Colonies on neutral Raulin with 10 per cent gelatin in petri dishes, thin and flexuous, slowly growing, becoming 2.5 to 3 cm. diameter in twelve days, at about which time the gelatine quickly liquefies; surface velvety, for the most part superficially zonate, becoming convex with the marginal areas slightly or considerably raised, radiately wrinkled, with center also elevated, with the whole surface sprinkled or covered with white areas suggesting the residues of evanescent drops, margin loose, 0.5 to 1 mm. or even 2 mm., wide in the growing colony; in color at first pale green (at fruiting margin) C.d.C. 341, 342, 337, 338, becoming 289 to 314 or similar shades in age; reverse more or less zonate pale yellow such as 221, 171, 196; drops numerous uncolored, arising in the margin of the young colony; odor none; conidiophores about 300 or 400 by 3.5 to 4 $\mu$ , simple or rarely branched, with walls smooth or rough; penicillus long, usually asymmetrically arranged and 40 to 50 $\mu$  long; branches about 16 to 26 by 3 to 4 $\mu$ , with walls occasionally roughened, in groups of 2 or rarely 3; metulae about 12 to 15 by 2.5 to 3.5 $\mu$  in groups of 3 to 5; sterigmata about 9 to 10 by 2 to 2.5 $\mu$ , in verticils of 5 to 10 $\mu$  conidia 2.5 to 3.5 by 2.5 to 3 $\mu$ , smooth, variously ovate to subglobose, long adhering in masses.

Habitat: Species isolated from earth in pine woods on the ridge "Poronice" in the mountains "Tatry" in Poland. Zaleski classes it "Hemiconcentrica 1° non-inflata b. aspera." Our notes follow: Type strain growing poorly at or above 30°C.; at temperatures of about 20° to 25°C., colonies upon Czapek's solution agar, becoming 30 to 35 mm., in diameter in fourteen days, velvety plane or with a zone line 5 mm., from the edge, dull green or olive green or pale olive becoming shades of vinaceous in old cultures, with a white margin 0.5 to 1 mm. in width; on wort, colonies more or less floccose, buckled, convoluted and radiately wrinkled, and showing prominent marginal stolons, extending beyond the submerged mycelium and starting new submerged hyphae (as in

*P. stoloniferum* Thom); reverse pale yellowish; drops, colorless; conidiophores 3.5 to 4 $\mu$  in diameter; penicillus with the compact base of *P. stoloniferum* or *P. brevi-compactum*, consisting of a main axis and one verticil of metulae or a main axis, one branch and a second verticil of metulae forming a compact conidia producing mass with chains of conidia diverging; metulae 10 to 12 $\mu$  long; sterigmata 8 to 9 $\mu$  long; conidia about 3 $\mu$  in diameter.

Culture no. 5010.8 received from Baarn in July, 1928, appears to be type.

192. *P. szaferi* Zaleski. In Bul. Acad. Polionaise Sci.: Math. et Nat. Ser. B, 1927, pp. 447-448, Taf. 38. Zaleski no. 1048.

Colonies on neutral Raulin's 10 per cent gelatine, slowly growing, becoming 2.5 to 3 cm. in diameter in twelve days, with surface wrinkled or undulate and gelatine strongly liquefied, appearance velutinous or velvety, with crowded but indistinct zones, radiately wrinkled, whole surface hirsute with secondary mycelium, with central area elevated, margin (fimbria) 1 to 2 mm. wide when young, indefinite in age; in color pale green at first, C.d.C. 342, 343, to very dark yellowish green, an olive shade such as 269, 274, 148, 140; reverse and liquefied gelatine in orange, yellow tints C.d.O. 151, 157, conidiophores commonly 300 to 500 by 4 to 4.5 $\mu$ , straight or slightly flexuous with walls slightly asperulate; penicillus 40 to 50 occasionally 60 $\mu$  long, with walls mostly smooth; branches (rami) about 18 to 30 by 3.5 to 4 $\mu$ , asymmetrically arranged in groups of two or three, straight or slightly incurved with apices inflated or vesicle-like up to 4.5 $\mu$  in diameter; with walls somewhat asperulate; metulae about 12 to 14 by 3.5 to 4 $\mu$ , commonly somewhat enlarged and rounded at the apex and 4 to 6 in the group; sterigmata 9 to 10 by 2.2 to 2.5 $\mu$  straight, cylindrical, crowded in verticils of 8 to 12; conidia subglobose to globose, 2.5 to 3 or even 3.5 $\mu$ , smooth, long adherent in masses and showing connectives between members of the chains.

Habitat: Species isolated from earth under pine trees in "Długe Goslina" near Poznan in Poland. It was recognized by Zaleski as a member of Biourge's series *P. brevi-compactum* hence classed "*Hemiconcentrica* 1° noniflata a laevia."

Zaleski's description gives the sterigmata as remarkably long (insigniter proceris) but his measurements do not justify his phrase.

Our notes follow: Colonies upon Czapek's solution agar velvety at least toward the margin, more or less radiately wrinkled, yellowish

green (near Lincoln Green Ridgway XLI); reverse pale yellow to ochraceous shades; drops, colorless; conidiophores  $125\mu$  long, 4 to  $5\mu$  in diameter, with walls smooth; penicilli brevi-compactum type,  $35\mu$  in length; metulae about  $15\mu$  long; sterigmata 9 to  $10\mu$  long; conidia 2.5 to  $3.5\mu$  in diameter.

Culture no. 5010.24 received as type from Baarn in July, 1928, fully complies with the description and justifies its assignment to the group with *P. brevi-compactum* Dierckx.

*Conidia elliptical. Colonies fairly deep, tending to floccosity.*

193. *P. tabascens* Westling. Arkiv för Botanik 11, pp. 56, 100-102; figs. 20, 61. 1911.

Colonies in prune gelatine, floccose, with conidial areas blue-green (C.d.C. 363) then green (C.d.C. 317, 313, 338, 334, 309) and finally yellow green (C.d.C. 284, 285, 289) with a narrow white somewhat wooly margin during the growing period, and producing stolon-like hyphae at the margin reentering the substratum often beyond the submerged hyphae, with the production of various groups of terminal branches which become vegetative hyphae again, also marginal prostrate coremium-like bundles of hyphae appear; small drops of fluid appear among the conidiophores; reverse uncolored or pale yellow; gelatine quickly liquefied, with an alkaline reaction; odor slight or wanting; conidiophores arising from creeping hyphae, smooth, up to 1 mm. long by 3.8 to  $6\mu$ , often branched below, differentiated from the vegetative hyphae by their greater diameter; penicillus not over  $90\mu$  long, usually about as broad as long; metulae 11.5 to 15 by 3.2 to  $6\mu$ ; sterigmata 7.5 to 9 by 1.6 to  $2.5\mu$ ; conidia oval to globose or subglobose, smooth or delicately roughened mostly 2.5 to  $3\mu$ , exceptionally up to 4 or  $4.5\mu$ , in diameter.

Species found first in dried *Sagina* specimens, then in seeds of *Corylus avellaneus*. Westling recognized its close relationship to *P. stoloniferum* Thom from which he separated it on account of its globose conidia and its colony color which became more yellow green in age. In Westling's media growth at  $30^{\circ}$  to  $31^{\circ}\text{C}$ . was poor, in most of his media normal colonies were reported. Westling's culture was compared with *P. stoloniferum* and certainly belongs with it. Whether the separation of this form upon the differences noted is warranted is perhaps debatable.

Biourge Monogr. p. 189, gives a Latinized diagnosis drawn in his

own terminology from Westling's description but did not recognize the species in culture.

194. *P. aurantio-griseum* Dierckx var. *Poznaniensis* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 444-445, Taf. 37, Zaleski's no. 1306a.

Colonies on neutralized Raulin with 10 per cent gelatine in petri dishes, moderately coarse, more or less wrinkled, slowly growing, becoming about 3 cm. in diameter in 12 days, with gelatine partly but not completely liquefied; velvety or slightly subfloccose, distinctly or ridged zonate, more or less radiately wrinkled or undulate, with over-growth in center consisting of loose aerial hyphae sordid white and often extending to cover a major part of the colony becoming abundant at times toward the margin; central area raised; margin (fimbria) 1 to 1.5 mm. wide during the growing period; in color green C.d.C. 342, 343, 319; reverse in pale yellowish shades smooth, with radiate wrinkles showing, and more or less definitely zonate; drops abundant sometimes uncolored, again pale yellowish, scattered over the whole surface or more abundant at the marginal areas; conidiophore straight or more or less flexuous, commonly 600 to 800 $\mu$  (even 1200 $\mu$ ) long and 4.5 to 6 $\mu$  in diameter, with all walls smooth, penicillus simple and 35 to 50 $\mu$  long or branching and 60 to 75 $\mu$  long; branches commonly at 1 level, occasionally with a second verticillate series bearing the metulae figured as 1 divergent branch bearing a dense cluster of secondary branches, below the metulae; metulae with apices usually enlarged, often clavate or capitate; sterigmata straight, fairly long, with short tubes, about 10 to 11 by 2.5 to 2.8 $\mu$ , commonly 6 to 8 in the verticil; conidia smooth, mostly subglobose, mostly 2.5 to 3 $\mu$ .

Habitat: Variety was isolated from earth in pine woods, known as "Lasy Trezebawskie" near Poznan in Poland. It differs from Biourge's interpretation of Dierckx's species in having smaller conidia, more numerous sterigmata, coarser conidiophores, differences in color. Zaleski puts it in "*Euconcentrica-classica*." Our notes follow: Colonies upon Czapek's solution agar growing well about 20°C., but poorly at 30°C., or above; at 20°C., almost velvety in appearance but with a basal felt of aerial hyphae, plane or radiately wrinkled, usually buckled in center, bluish green (light dull glaucous blue, Ridgeway XLVIII); faintly zonate in the outer areas in age (azonate in young colonies—hemizonate of Biourge), showing marginal "stolons" when grown in petri dishes of wort agar; reverse in yellow shades becoming buff in

age; odor none; drops not seen; conidiophores 100 to 300 $\mu$  by 3 to 3.5 $\mu$ ; penicilli with the compact branching system of Thom's *P. stoloniferum* or *P. brevi-compactum* Dierckx, and occasionally producing penicilli on short perpendicular branches of the conidiophores; conidia elliptical about 3 by 2 to 2.5 $\mu$ .

Culture no. 5010.2 received from Baarn in July, 1928, as the type of this species is probably correctly named although Zaleski's figures show a triverticillate penicillus more consistently than our transfers.

*Conidia showing persistent ellipticity. Colonies tending toward velvety.*

195. *P. griseo-brunneum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88. 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 162-163; Col. Pl. II, Cart. 148; Pl. III, fig. 19. 1923.

Colonies on wort gelatine hemizonate, radiately wrinkled, blue green to green (C.d.C. 338), in age sordid brown, with white marginal zone 2 mm. broad, undulate wrinkled, producing sessile coremia, like cabbage heads or cauliflower; reverse bluish green, to yellowish; odor none; drops amber; conidiophore 4.5 to 5 in diameter, long, smooth; penicillus 40 to 60 $\mu$  long, figured as biverticillate or with main axis and one or two branches each producing a compact verticil of metulae; metulae 10 to 15 by 2 to 4 $\mu$ , in verticils of 2 to 5; sterigmata 7.5 to 12 by 2.5 to 3 $\mu$ ; conidia subglobose 3 to 4 by 2.5 to 3 $\mu$ , with connective.

Biourge's no. 148 (our 4733.68) is apparently type: Colonies upon Czapek's solution agar spreading up to 5 to 6 cm., in diameter in two weeks, velvety or nearly so about 200 to 300 $\mu$  deep, radiately wrinkled, showing traces of zonation toward the center but not at the growing margin, again showing more or less marginal zonation; in color bluish gray green shading to green and later to slate olive beginning in the central area; on gelatin plates, stoloniferous at the margin; reverse in yellow shades ("clay yellow") to "cream buff" (Ridgway) or "pinkish buff" in older areas; drops colorless to yellowish; conidiophores about 100 to 200 $\mu$  by 2.5 to 3 $\mu$ , with walls smooth; penicillus consisting of a main axis and one or a cluster of branches with metulae, 10, 12, or even 18 $\mu$  long and sterigmata 7 to 8 $\mu$  long; conidia elliptical about 4 by 2.5 to 3 $\mu$ , or in other cultures partly approaching subglobose.

Another culture (no. 5016.9) isolated from a rotting *Strobilomyces* is apparently nearly related to Biourge's species.

196. *P. patris-mei* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 496, 497, 498; Taf. 58; Zaleski no. 1420.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 26 to 30 $\mu$  in diameter in twelve days, liquefying the gelatine only slowly and partially, velvety or slightly floccose, azonate or with a trace of zonation near the margin, whole central area depressed, thrown into cerebriform wrinkles, irregular in outline with a marginal pale yellowish band or zone; in color, for several days, white, becoming orange yellow and yellow shades such as 178A, 203A in the marginal zone and slowly showing indistinct green shades such as 322 in areas within; reverse and liquefied gelatine in orange-yellow and orange shades 171, 166, 104, 132; odor none; conidiophores about 50, 100 to 30 or up to 600 $\mu$  by 2 to 2.5 $\mu$ , commonly somewhat enlarged at apex, unbranched, erect or ascending, flexuous; penicilli 8 to 10 $\mu$  long when simple, 15 to 25 $\mu$  when branched, with all walls smooth; metulae occasionally present about 8, 10 to 16, or 18 by 2 to 2.5 $\mu$ , commonly somewhat enlarged at the apex, unequal in length and asymmetrically arranged; sterigmata about 7 to 8, by 2 to 2.3 $\mu$ , in verticils of 3, 6 to 15, or 20, or occasionally occurring singly, with tubes usually small and short; conidia 2 to 2.5 $\mu$ , smooth, subglobose to globose, with connectives evident.

Habitat: Species isolated from earth under pine woods in square 652 of the forest Puszcza Bialowieska in Poland.

Zaleski places it in "*Aspergilloides* Wehmer-Dierckx, Series B, candido-fulvum Dierckx." Our notes follow: Type strain growing well at 20°C., poorly at 30°C., and above; colonies upon Czapek's solution agar about 20°C., velvety or somewhat floccose in central areas, buckled and radiately wrinkled; green with a trace of yellowish (*artemisia* green Ridgway XLVII) becoming purplish brown in age; reverse pale yellow to grayish brown; odor, none; drops slightly yellowish, rather small, well distributed; penicilli of *P. brevi-compactum* type, with main axis up to 20 $\mu$  and the branch sometimes shorter, sometimes longer—up to 30 $\mu$  in length; metulae 10 to 14 $\mu$  long, in verticils compact especially at base; sterigmata 8 to 10 $\mu$  long; conidia more or less elliptical, apiculate when young, up to 3.5 $\mu$  in long axis.

Culture no. 5010.29 received as type from Baarn in July, 1928, appears to satisfy Zaleski's description.

197. *P. Bialowiezense* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 450, 451, Taf. 39; Zaleski no. 1597a.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes,

slowly growing, becoming 2.5 to 3 cm. in diameter in twelve days, with liquefaction of the gelatine beginning about the tenth day and progressing rapidly, thin, plane, velvety, distinctly zonate only in the outer areas; central areas becoming convex showing the marks and dried residues of drops, also showing a central elevation or umbilicus (?) and some overgrowth of sordid white aerial mycelium; margin 2 mm. wide at first then 1 mm. during the remainder of the growing period; in color pale blue green at first C.d.C. 371, 372, then green, 343, reverse and liquefied gelatine in pale yellow, 166, 156, 171, and related tints; drops few uncolored arising near the marginal area; odor none; conidiophores more or less straight, 400 to 600 by 4 to 5 $\mu$ , penicillus mostly 60 to 70 $\mu$  long, with branches mostly at three levels occasionally four; basal branches when present 30 to 35 by 4 to 5 $\mu$ , the next series 12 to 20 by 3 to 4 $\mu$ , with walls delicately roughened; metulae about 10 to 12 by 3 to 4 $\mu$  in groups of about 4 to 6, enlarging upward (clavate); sterigmata about 10 to 11 by 2.5 to 3 $\mu$ , in verticils of 5 to 8; conidia smooth, mostly ovate, some subglobose 2.5 to 3.5 by 2.5 to 3 $\mu$ , with connective evident between members of the chains.

**Habitat:** Species isolated from earth under coniferous trees at about 8 cm. depth in square "369" of the forest called "Puszcza Bialowieska."

Biourge regarded this as related to *P. brevi-compactum* Dierckx from which it is separated by its rough cell walls. Zaleski classes it as "Hemiconcentrica 1° non-inflata b. aspera." Our notes follow: Type strain upon Czapek's solution agar growing well at 20°C., but only slightly at 30°C., or above; colonies spreading slowly, velvety with or without a cottony overgrowth in age, plane or slightly elevated in center, gray green to bluish green, becoming brown in several weeks; reverse in shades of yellow, orange, yellowish brown, fawn or avellaneous, at various stages of growth; drops pale yellowish brown, abundant; conidiophores up to 600 $\mu$  long, or longer, by 4 to 5 $\mu$ , with walls faintly pitted or granular, especially toward the apex; penicillus with primary branches up to 30 or 35 $\mu$  long, commonly with a secondary clavate series 10 to 12 $\mu$  in length bearing the metulae making a triverticillate mass up to 70 $\mu$  long, asymmetrical and closely compacted at base (*P. brevi-compactum*-like), with walls all delicately pitted or granular in appearance; metulae 10 to 12 $\mu$  in length; sterigmata 10 to 12 $\mu$  long; conidia elliptical 3 to 3.5 by 2 to 2.5 $\mu$  in loosely diverging or coiling chains.

Culture no. 5010.4 from Baarn appears to be Zaleski's type.

## CHAPTER XVI

### THE LANATA-TYPICA

#### ASYMMETRICA

*Section 3. Lanata-typica* (Lanata of Biourge emended): In this section of the Asymmetrica colonies show a definitely vegetative aerial mycelium consisting of a cottony, lanose or floccose mass, web or felt from which conidiophores arise as branches (not giving the velvety appearance). Conidial areas commonly appear centrally after the establishment of the definite aerial felt and progress toward the marginal areas in which the aerial felt in some species disappears toward the end

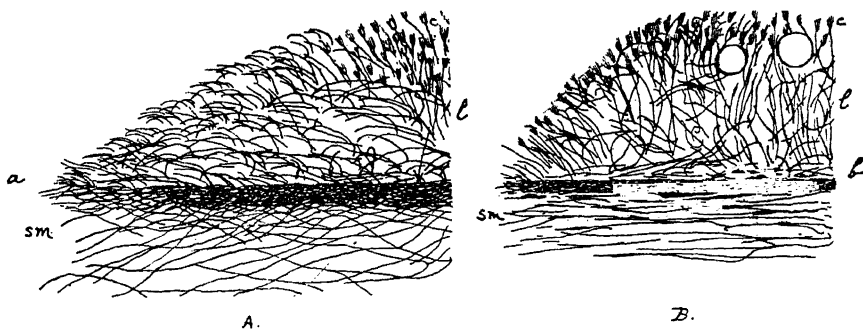


FIG. 43. *P. caseicolum* A, and *P. camemberti* B.: Diagrammatic radial sections (magnified 25 times) showing: *ab*, the agar line; *sm*, submerged mycelium; *l*, the floccose mass of vegetative hyphae; *c*, conidiophores and penicilli in their relation to floccose hyphae.

of the growing period leaving the margin velvety. Conidial chains usually become a tangled mass when ripe rather than forming columnar masses (fig. 43).

Biourge (Monogr, p. 29) made *P. camemberti* Thom the type of his Sub-section III Lanata, adding with it the species recognized here as *P. caseicolum* Bainier and *P. biforme* Thom. We have included a number of species placed by Biourge in his Zonata believing that the floccose aerial felt as a character brings together more nearly related species than the formation of zones in the colony grown as Biourge grew it. As used here the line drawn is still arbitrary since border-line forms between the



floccose section of the monoverticillata and the floccose asymmetrica can be placed in either section without straining the imagination. In fact these sections merge insensibly into each other and forms are simply placed by the judgment of the observer as to the place in which they may be most easily identified. Species preponderatingly floccose but showing trailing or ascending bundles of hyphae have been put among the Lanata-funiculosa (Chapter XVIII). The floccose type of colony merges into the velutinous type of colony in Biourge's Hemizonta characterized by *P. brevi-compactum* Dierckx (*P. stoloniferum* Thom) with its very characteristic type of penicillus and in Biourge's Radiata (*P. chrysogenum* group) which maintain for the most part a beautifully velvety surface above a basal felt of mycelium.

The sub-sections proposed are frankly arbitrary places to put the species into some sort of order.

*Colonies with developing conidial areas in shades of ochraceous, traces of green if present transient or evanescent.*

This section includes some series of closely related species and others more or less widely divergent. The arrangement offered is proposed mainly for convenience in identification although real relationship is indicated as far as possible.

*Key to lines of separation used*

- I. Colonies showing some shades of green with ripening conidia.....III.
- I. Species lacking green color in ripe conidia.....II.
- II. Colonies in shades of yellow, orange yellow, ochraceous, olive to reddish brown in age.....*P. ochraceum* and allies, 200.
- II. Colonies in pink or rosy shades with ripening conidia, show the colony aspect of Gliocladium and conidia long elliptical.....*P. vermoeseni*, 451.
- II. Colonies white or occasionally pale cream.....*P. caseicolum* and allied strains.
- IIa. Synonyms.....*P. caseicolum* Bainier, 202.
- IIb. ....*P. camemberti* var. *rogeri* Thom.
- IIc. ....Type Thom No. 6.
- IId. ....*P. camemberti* of Roger and Maze.

- IIe. .... *P. candidum* Roger-  
fide Biourge, 203.
- IIf. .... *P. candidum* Link.
- III. Colonies showing some shade of green in ripening  
conidia, which may become gray, olive, or  
brown in age. .... IV.
- IV. Colonies zonate definitely at least in the latter  
part of growing period. .... XX.
- IV. Colonies azonate or rarely showing traces of  
zonation in age. .... V.
- V. Ripe conidia globose or subglobose. .... VI.
- V. Ripe conidia elliptical persistently. .... XIV.
- VI. Ripe conidia 4 to 5 or even 6 $\mu$  in diameter. .... VII.
- VI. Ripe conidia globose or subglobose 4 $\mu$  or less in  
diameter. .... XI.
- VII. Reverse colorless. .... VIII.
- VII. Reverse colorless then slowly yellow. .... IX.
- VII. Reverse orange yellow. .... X.
- VIII. Colonies slowly pale green. .... *P. camemberti* Thom,  
205.  
Type culture Thom  
No. 5.
- Synonyms or possible synonyms. VIIIa. .... *P. album* Epstein.  
VIIIb. .... *P. Epsteinii* Lindau,  
204.  
VIIIc. .... *P. album* Preuss.  
VIIIId. .... *P. camembert* Sopp,  
206.  
VIIIE. .... 4291.19 Hanzawa.
- IX. Reverse colorless then slowly yellow or pale  
yellow. ....
- IXa. Reverse colonies in yellow shades. .... *P. lanoso-viride*  
Thom, 203.
- IXb. Colonies dull gray green. .... *P. flavido-margina-*  
tum Biourge, 209.
- X. Reverse orange yellow.
- Xa. Colonies with mycelium white then yellowish  
orange and slowly developing conidial areas  
bluish green. Conidia partly subglobose,  
partly elliptical. .... *P. aurantio-vire*  
Biourge, 210.

- XI. Ripe conidia globose or subglobose less than  $4\mu$   
in diameter.....XII.
- XI. Ripe conidia mostly elliptical (occasionally sub-  
globose).....XIII.
- XII. Colonies in green or gray green shades.....*P. lanosum* Westling,  
211.
- XII.....*P. Raciborskii* Zal.,  
212.
- XIII. Conidia more or less persistently elliptical  
(occasionally subglobose).....XIV.
- XIV. Floccose mycelium uncolored.....XV.
- XIV. Floccose mycelium orange next the substratum;  
reverse orange to bay.....*P. aurantio-candi-*  
*dum* Dierckx, 213.
- XVa. Conidial areas green to gray green.....*P. biforme* Thom,  
215.
- XVb. Conidial areas scanty grayish or bluish.....*P. aurantio albidum*  
Biourge, 216.
- XVc. Conidial areas green or gray green, with reverse  
in spots areas or zones becoming reddish  
brown.....*P. janthogenum* Bi-  
ourge. See Fasci-  
culata.
- XVd. Conidial areas abundant blue green, reverse  
uncolored; floccose masses deep.....*P. lanoso-coeruleum*  
Thom, 217.
- XX. Colonies with zonation well marked at least in  
the latter part of the growing period.....XXI.
- XXI. Conidia elliptical.....XXV.
- XXI. Conidia globose.....XXII.
- XXII. Drops yellow.....XXIII.
- XXII. Drops uncolored or not conspicuously colored....XXIV.
- XXIII. Colonies bluish green to gray green; reverse  
yellow to reddish.....*P. roseo-citreum* Bi-  
ourge, 218.
- XXIVa. Colonies blue green; reverse more or less  
yellow.....*P. solitum* Westling,

- XXIVb. Colonies with a trace of bluish green at margin, quickly dull green or gray green, walls smooth or slightly pitted or rough forming a felt 300 to 500 $\mu$  deep.....*P. commune* Thom, 219.
- XXIVc. Colonies gray green with walls markedly rough forming a felt often 500 to 800 $\mu$  deep.....*P. fusco-glaucum* Biourge, 220.
- XXIVd. Colonies gray green forming felts 2-3 mm. deep in center thinning at margin; reverse yellow to reddish orange.....*P. griseo-fulvum* Biourge, 222.
- XXIVe. Colonies in bright green shades; funiculose overgrowths tardily showing.....*P. psittacinum* Thom, 270.
- XXV. Colonies in gray green to gray shades deeply floccose reverse colorless; conidia 4 by 3 $\mu$ .....*P. lanoso-griseum* Thom, 221.

200. *P. ochraceum* (Bainier nomen nudum) Thom.

Colonies on Czapek's solution agar forming closely felted aerial masses 300 to 500 $\mu$  deep with a narrow white border at the agar level and a central area variously wrinkled and buckled into masses of agar and submerged mycelium up to 2 to 3 mm. deep, azonate or indistinctly zonate at first but becoming definitely zonate at the margin especially in age, with newer central areas faintly greenish yellow or olive (near R. XXXI. Sea foam yellow passing to olive-buff R. XL) and related shades and finally to dark vinaceous brown (R. XXXIX) or related shades in age (several weeks usually); reverse colorless at first becoming variously yellowish, or pinkish, ochraceous or vinaceous brown shades under various conditions; conidiophores arising either from aerial hyphae or from submerged hyphae 100 to 200 $\mu$  or longer, by about 4 $\mu$  in diameter with walls abundantly pitted and often warty; penicillus consisting of branches from one or more of the upper nodes, often slightly diverging, with metulae, sterigmata, and diverging or tangled chains of conidia; branches 15 to 25 $\mu$  long; metulae 8 to 10 $\mu$  long; pitted walls evident on branches and metulae but not on sterigmata; sterigmata 7 to 8 by 1.5 to 2 $\mu$ ; conidia subglobose about 3 $\mu$  (up to 3.5 $\mu$  occasionally) smooth or on very careful examination showing a suggestion of pitting.

Type 4640.449 received from the Bainier collection through Dr. Fonseca as a labeled tube and described here.

**201. *P. ochraceum* var. *macrosporum* n. var.**

Colonies varying from the type in the reduction or entire suppression of the greenish shade in conidial areas leaving colonies mostly olive or olive gray (R. Plate LI.) Not reaching the deep vinaceous brown shades of the species; conidia somewhat larger about  $4\mu$  or in one strain reaching  $4.5\mu$  at times.

Cultures 4424 from corn (*zea mays*) collected in Cleveland, Ohio and no. 4742P<sub>3</sub> contributed by H. R. Jones from the studies of moldy tobacco at the University of Bristol in England.

*Colonies lacking ochraceous shades in their developing conidial areas. Colonies white or pale cream.*

**202. *P. caseicolum* Bainier, Bul. Soc. Mycol. France 23: 94, Pl. X., fig. 6-10. 1907. (See fig. 43, 44.)**

Synonym: *P. camemberti*, var. *Rogeri* Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, p. 52-53, fig. 17, 1910; *P. candidum* of Roger and Mazé, not Link; *P. epsteinii* Lindau, q. v.; *P. rogeri* Wehmer in Lafar Tech. Myk. 2 Aufl. 4 p. 226.

Colonies grown upon sugar gelatin or bean or potato agar pure white, in age somewhat cream at times, loosely and evenly floccose to the very margin, where aerial and submerged hyphae grow with equal rapidity. Reverse of colonies white or yellowish white (not discolored); conidiophores 3 to 5 by 100 to  $800\mu$  varying greatly, mostly branches of aerial hyphae; penicillus 70 to  $90\mu$  in length, loosely and irregularly branched and bearing rather few metulae at unequal heights, with divergent chains of colorless conidia; branching system of conidial fructification sometimes  $75\mu$  in length; conidia smooth, hyaline, 4 to 4.5 or even  $5.5\mu$  in diameter, globose or nearly so when ripe; sugar gelatin slowly liquefied under the center of the colony only, colonies never floating in a pool of liquid; reaction in the medium acid to litmus at first, then changing to alkaline.

This fungus has been found by us only upon Camembert, Brie, and Neufchatel cheeses from western Europe. Our no. 4640.440 from the Bainier Collection may justly be regarded as type. It was discussed by Mazé as *P. candidum* Link. This seems an impossible application of the name *P. candidum* from Link's description or that given by Saccardo, since the spores are stated to be 2 to  $3\mu$  in diameter. Further, in such identifications no account is taken of a paper by Morini, in which an ascigerous stage is described for *P. candidum* Link. Under many years of cultivation no signs of an ascigerous form have been produced. Stoll

has considered *P. candidum* to be only a colorless *P. glaucum*, but as the author has so far failed to find a worker who will undertake to limit the name *P. glaucum* to a special form, this does not mend matters. Long cultivation does show, however, that this organism is closely related to the one already described as *P. camemberti*. Since this is the form given prominence in cheese studies by the work of Georges Roger, it seemed most natural to regard it as a variety of the former species and designate it by Roger's name.

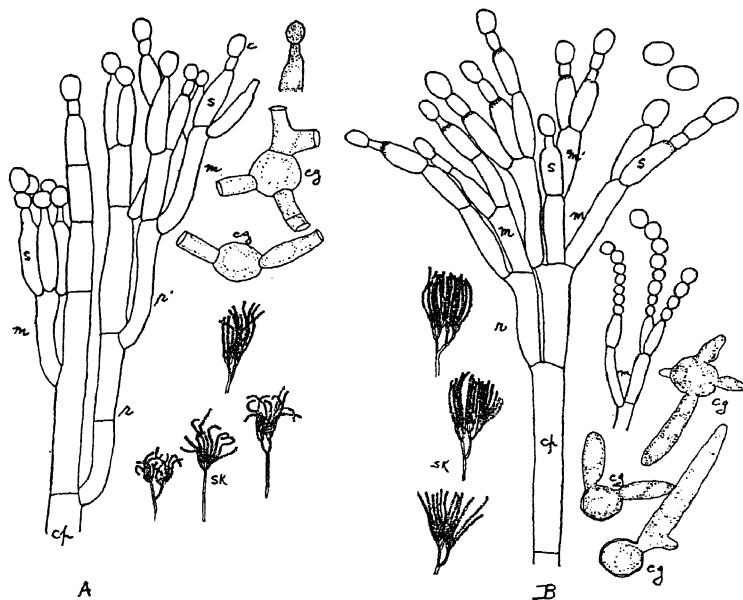


FIG. 44. *P. caseicolum* A, and *P. camemberti* B: c, conidia; cg, conidia germinating; cp, conidiophore; r, branches or rami; m, metulae; m', secondary metula in verticil with sterigmata; sk, habit sketches.

In Lafar's *Handbuch der Technischen Mykologie*, Professors Weigmann and Wehmer refer to this fungus as probably identical with *P. camemberti*. Although this might seem possible from examination of the literature alone, no one actually familiar with the cultures will claim such identity.

Bainier's figures and descriptions establish the identity of *P. caseicolum* with *P. camemberti* var. *Rogeri* Thom, and *P. candidum* of Mazé and

of Roger, and *P. candidum* Roger in Biourge Monogr., p. 193, Pl. V, fig. 27 and Biourge's culture no. 4733.26. This has been used in making Brie and Camembert cheeses under Roger's patents in certain regions of France. While the texture and to some degree the flavor of cheese made with this species (see Chapter IX) differs from the usual type of Camembert cheese, the organisms are so nearly identical in all but color that Thom (1910) preferred to regard this form as a variety of *P. camemberti* (Thom 1910), until its identity with Bainier's species was fully established.

3. *P. candidum* Roger. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 193-194; Col. Pl. III, Cart. 157; Pl. V, fig. 27. 1923, was cited from Roger (Revue Hebdomadaire (Paris) 7: 334), but Roger regarded his form as *P. candidum* Link.

As synonymy Biourge gives *P. camemberti* var. *rogeri* Thom, *P. caseicolum* Bainier, *P. album* Epstein nec Preuss; *P. epsteinii* Lindau.

Biourge's culture no. 157 (our no. 4733.26) is apparently identical with Thom's no. 6. *P. camemberti* var. *rogeri* Thom. Biourge is hardly justified in following Roger by discarding Link's *P. candidum* and attributing the species to Roger even though we know that Roger used this species under his patent in ripening cheese. Roger's description is not adequate; his organism was obtainable only by isolation from cheeses made by his method or by purchase of his culture.

204. *P. epsteinii* Lindau. Deutsch. Krypt. Flora, Pilze 8: 166.

Synonym: *P. album* in Epstein, Arch. f. Hyg. 45: 360. 1902.  
Probably *P. caseicolum* Bainier.

This is a manuscript species only since Lindau noting the previous use of the specific name *P. album*, collected a few items of morphology making a totally inadequate description, out of Epstein's discussion of this mold and its activity in cheese and proposed a new specific name for it. It may be dropped.

*Colonies showing some shade of green with the ripening of conidia.*

205. *Penicillium camemberti* Thom. Emended from U. S. Department of Agriculture, Bureau of Animal Industry, Bul. 82, p. 33, fig. 1, 1906; also Bul. 118: 50, fig. 16, 1910. (See Fig. 43 and 44.)  
Possible synonym: *P. album* Epstein (not Preuss), Archiv. f. Hyg.,

Bd. 45, Hft. 4, p. 360, 1902. *P. epsteinii* Lindau, Deutschl. Krypt. Flora, Pilze, VIII, p. 166. *P. camembert* Sopp.

See also Biourge La Cellule 33: fasc. 1, pp. 191–193; Col. Pl. III, (Biourge gives VIII incorrectly); carton 5; Pl. V, fig. 26, 1923; and Westling Arkiv för Botanik 11: pp. 73–74, fig. 7, and 44, 1911.

Colonies on potato agar or lactose gelatin effused; white (sometimes yellowish white), changing in 5 to 8 days to pale gray-green (glaucous); surface of colony floccose, of loosely felted hyphae about  $5\mu$  in diameter, reverse of colony yellowish white; conidiophores 300 to  $800\mu$  in length, 3 to  $4\mu$  in diameter, septate, cells thin-walled, walls delicately pitted or marked, often collapsing in age, arising as branches of aerial hyphae; penicillus sometimes  $175\mu$  in length, but usually much less, consisting commonly of one main branch and one lateral branch, sparingly branched to produce rather few sterigmata which bear long loosely divergent chains of conidia; sterigmata 8 to 11 by 2.4 to  $3\mu$ ; conidia at first cylindrical, then elliptical, and finally globose when ripe, smooth bluish green by transmitted light, thin-walled and commonly guttulate, 4.5 to  $5.5\mu$  in diameter, swelling in germination to 8 to  $10\mu$ ; germ tubes one to several; cells of mycelium about 5 by 20 to  $40\mu$ ; liquefies lactose gelatin only under center of colony; produces a strong alkaline reaction in gelatin, free from sugar, but in sugar media produces a more or less persistent acid reaction; growing and fruiting period, about two weeks; fruits only upon exposed surface of the substrata; never produces spores in cavities not broadly open.

Habitat: Camembert and other soft cheeses.

206. *P. camembert* Sopp. Monogr. pp. 179–180. Taf. XIX, fig. 134; Taf. XXIII, fig. 17–18, 1912.

Synonym: *P. aromaticum* III. Sopp; *P. album* after Epstein not Preuss; *P. camemberti* Thom no. 205.

Sopp adds nothing to Thom's description of this cheese mold.

207. *P. album camemberti*.

Listed as no. 601 in Catalogue of the National Collection of Type Cultures, Lister Institute, London, p. 26, 1922, as contributed in 1920 by Thaysen who obtained it from Liebefeld, Berne, Switzerland. Undoubtedly this is an incorrect form for *P. camemberti* Thom. In one reference this combination is ascribed to J. Arthaud-Berthet but the original of this usage has not been found.



The Camembert cheese mold is about as tangible a species of mold as we possess. It was probably isolated by Sopp, by Epstein, by Roger, by Mazé and his co-workers, certainly by some of them before Thom published his description. Ripe cheese with the qualities imparted by this species has been produced for a long time in northern France. So long as the industry was restricted to the homogeneous region where it developed, the problems of propagation and control were not significant. With attempts at the development of Camembert manufacture in other regions of France and in Germany investigations were necessary to overcome the handicaps of infection by other organisms in factories subject to different climatic conditions. These difficulties were greater when the task of establishing such manufacture was attempted in America. Thom purchased the higher grades of imported Camembert, found this species to be the dominant organism present upon their surface and demonstrated its function in cheese ripening hence the description and name applied (see Chapter IX).

The species is easily grown in pure culture upon a wide range of culture media. Its habitat in nature outside the cheese industry was not determined since no strains have been encountered which could be traced definitely to other sources than the dairy manufacturing industries. It is certainly uncommon as a mold upon other substrata.

*Reverse colorless then slowly yellowish to drab in age.*

**208. *P. lanoso-viride* Thom, n. sp.**

Colonies upon Czapek's solution agar azonate, floccose about 1 mm. deep, slowly spreading over the substratum, with marginal area broad white during the growing period, with conidia area in rather bright green shades such as "water green" "Pois green" or "grape green" of Ridgway Plates XLI and XLVI (near C.d.C. 337, 338), fading unevenly in age to light shades of olive gray, reverse uncolored then tardily and unevenly becoming shades of drab; drops colorless, abundant rather small tending to develop in concentric lines but without accompanying signs of zonation; odor faint but penetrating; conidiophores up to 1 mm. by 4 to 5 $\mu$ , more or less sinuate or flexuous, with walls pitted or rough; penicilli about 50 $\mu$  long with walls pitted or rough, irregularly branched and bearing tangled chains of conidia; with metulae produced at different levels and variously duplicated by secondary metulae, producing sterigmata at several levels; both metulae and sterigmata tending to fall away in mounted preparations from older areas; branches 20 to 40 $\mu$  long, more

or less divergent; metulae (10) 15 to  $20\mu$  long with apex enlarged; sterigmata mostly 10 to 12 less commonly 14, 16 or  $18\mu$  long by 3 to  $4\mu$  in diameter, often tapering gradually to rather long tubes; conidia mostly about  $4\mu$  to  $4.5\mu$  in diameter but showing occasionally cells much larger, and more or less elliptical, smooth, thin walled, and homogeneous in appearance, slightly colored as seen under oil immersion.

Type (our no. 5034.12) received in the collection of Nobel's Explosives Company sent by J. H. Birkinshaw in January, 1929, as isolated from the "sweet waters" of a glycerine still at Ardeer, Scotland. The colonies suggest in color the bright green species described (no. 270) as *P. psittacinum* on account of its "parrot-green" color noted and described by Biourge. It differs from that species in lacking distinct zonation and in lacking the ropes of hyphae clearly evident at the edges and over the surface of the older colonies of *P. psittacinum*.

Dr. Raistrick's notes show a range in conidial diameter from 2.5 to  $7\mu$  and regard  $3\mu$  as average; gelatine either with or without glucose was not liquefied; colonies did not grow at  $37^{\circ}\text{C}.$ , but developed when culture was transferred to  $20^{\circ}\text{C}.$

*Colonies yellow in reverse.*

209. *P. flavido-marginatum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 150 to 152; Col. Pl. III, Cart 111; Pl. IV, fig. 24. 1923.

Colonies on wort gelatine with a trace ("minime") of zonation, plane; conidial areas at first pale blue (C.d.C. 422), with sterile margin 3 mm. wide in 2 zones, outer white, inner pale yellowish; coremia none; odor faint, that of moldy fruit; gelatine liquefied brownish red; conidiophore about  $3\mu$  in diameter; penicillus about  $60\mu$  long with all walls smooth, figured as variously branching and proliferating to produce sterigmata as several levels; branches in pairs 20 to 30 by 3 to  $4\mu$ ; metulae very irregular 7 to 12 to 14 even up to  $40\mu$ , by 2 to  $4\mu$ , in twos or threes; sterigmata 10 to 12 by 3.5 to  $4\mu$ , in twos, threes, or fours, often one or more in a verticil becoming secondary metulae; conidia 4 to 6 by 4 to  $5.5\mu$  ovate to subglobose.

Biourge's no. 111 (our no. 4733.50) on Czapek's solution agar radiate wrinkled, and more or less hemizonate in thin areas, azonate in deeper areas of the agar, thinning to almost velvety at the very margin in some cultures, definitely felted floccose in the deeper areas of the colony; reverse in Czapek tubes after 10 days purplish and almost black in center, 500 to  $600\mu$  deep at the broad white margin, and up to 1 mm. deep

toward the center, conidial areas dull glaucous gray green (Ridgway); reverse becoming pale yellowish especially in center; sometimes diffused; odor weakly moldy; drops colorless; penicillus approximating that of *P. camemberti*, with walls smooth; conidia 4 to  $4.5\mu$  mostly about  $4\mu$ .

The yellowish marginal zone used by Biourge as the basis of the name appeared in our notes only when tubes of this species were incubated to  $31^{\circ}$  to  $35^{\circ}\text{C}$ .

210. *P. aurantio-virens* Biourge. Monogr. La Cellule 33: pp. 119–121; Col. Pl. I, Cart. 77; Pl. I, fig. 5. 1923. Biourge suggests relationship to *P. pinophilum* Hedgcock, *P. lagerheimii* Westling, *P. aurantio-albidum* Biourge, and *Coremium citrinum* of Persoon's Myc. Europaea 1, p. 42.

Colonies in wort gelatine indefinitely zonate, plane, not wrinkled or buckled; conidial area green (C.d.C. 338, 339) or on rice and bean agar blue green; coremia occasional, with stalks yellow, atypical (?CT) found at the margin upon some media; reverse pale yellow to orange, to bay brown; odor indefinite; conidiophore 2.5 to  $3\mu$  in diameter, more or less flexuous; penicillus 60 to  $150\mu$  long, figured as main stalk and very long, appressed branches, more or less sinuate, and bearing metulae and sterigmata either at one level or more; occasional biverticillate penicilli are shown; branches 30 to  $40\mu$  or longer by  $2.8\mu$ ; metulae 11 to 12 by  $2.5\mu$ , 2 to 4 in the verticil; sterigmata 8 to 13 by 1.5 to  $2.5\mu$ , more or less irregular, and acuminate, two to four in the verticil; conidia 2.8 to  $4.8\mu$  by  $2.5$  to  $4\mu$ , ovate (subglobose rather than definitely elongated—C. T.).

Cultures Biourge no. 77 (our no. 4733.9): as received this culture gave the general reactions of the blue green series.

Colonies on Czapek's solution agar forming a rather broadly spreading floccose mass up to 500 to  $700\mu$  deep, at first white then yellowish orange near cream (between Saccardo's *cremeus* and *ochroleucus*) then becoming bluish gray green beginning in the central area; drops colorless; reverse orange yellow while the colony is colorless to cream, fading out as green color develops; odor present but not strong; conidiophores up to 300 to  $500\mu$  long by about 2.5 to  $3\mu$  in diameter, walls smooth or with the faintest traces of pitting; penicillus up to  $100\mu$  long or longer, compact, with conidial chains tangled; with appressed branches at different levels and varying in length in the same group from very short to  $40\mu$  long; sterigmata 10–12 by  $2\mu$ , borne at several levels of the penicillus; conidia dark green elliptical at first, becoming slowly more or less subglobose, varying in size up to about 4 by  $3.5\mu$  mostly with some persistent ellipticity.

From our own cultures we are inclined to believe Biourge's organism was not a pure culture but contained a submerged factor close to *P. expansum*. When freed from this contaminant we get the organism described here which may well carry the name with an emended description.

*Conidia less than  $4\mu$  in diameter.*

211. *P. lanosum* Westling. Arkiv för Botanik 11, pp. 55, 97-99, fig. 18, 60. 1911.

Colonies in prune gelatine, lanose, at first white then with the center becoming slowly gray green (C.d.C. 347, 318, 323, 343, 299), with a broad white margin, the whole often overgrown with white mycelium, becoming darker shades in age and finally dark brown; reverse uncolored or slightly yellow; gelatine slowly and partly liquefied in fourteen days, with an acid reaction when litmus is used; odor weak and scarcely definite; abundant drops appeared on the surface of the colonies; conidiophores smooth, up to 1 mm. by  $3.4$  to  $4.6\mu$ ; metulae 12 to 14 by 3 to  $4.6\mu$ ; sterigmata 7 to 9 by 2 to  $2.7\mu$ ; conidia globose, uniform, smooth or slightly roughened  $2.2$  to  $3\mu$  in diameter, swelling in germination to 5 to  $6\mu$ , in chains separating easily.

Westling's *P. lanosum* may be regarded as the type species of a series of floccose forms with conidia varying up to  $3\mu$  in diameter but not much larger; cultures of these species run through a series of shades of grayish green in the growing period toward brown shades in age. Several such strains have been seen from different sources.

A strain received from Nobel's Explosions Company in Scotland by courtesy of Mr. J. H. Birkinshaw and labeled *P. glaucum* satisfies Westling's description well enough to be assigned to *P. lanosum*. Our notes follow: Colonies upon Czapek's solution agar, floccose forming a mass or felt 1 to 2 mm. deep or occasionally deeper, becoming zonate in age, becoming broad white felts then developing from center outward conidial areas at first pale green, later such shades as "dark glaucous gray" of Ridgway XLVIII passing to gray shades such as "court gray," "Hathi gray" of Ridgway's table LII, or "mouse gray;" reverse colorless to fairly bright yellows under different conditions; odor not noticeable; drops colorless, few, distributed in the greenish areas; conidiophores up to 2 mm. long by about  $3\mu$  in diameter, smooth; penicillus with various but restricted branching and conidial chains at first parallel then loosely tangled; metulae 15 to  $20\mu$  long; sterigmata 8 to 10 by  $2\mu$ ; conidia about

3 $\mu$ , few up to 3.5 $\mu$ , with individual cells much larger (probably germinating), more or less persistent in chains when mounted.

Biourge (Monogr. La Cellule 33: fasc. 1, pp. 149; Col. Pl. XI, Cart. 357; Pl. XIX, fig. 109, 1923) describes his strain in terms which exclude it from Westling's species. This conclusion is confirmed by study of his culture.

212. *P. Raciborskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 454, 455; Taf. 36; Zaleski no. 249.

Colonies on neutral Raulin with 10 per cent gelatine, slowly growing becoming 3 to 3.5 cm. in diameter in twelve days, with gelatine liquefied, fairly coarse, lanose, not zonate, strongly and irregularly undulate; margin (fimbria) consisting of loose hyphae, 1 to 1.5 mm. wide during the growing period; in color becoming blue green, C.d.C. 368 in center at about six days with a broad white margin 3 to 5 mm. wide, with conidial areas becoming progressively 372, 347, 343, and finally dark orange brown shades in age such as 139, 140, 143; reverse and liquefied gelatine in pale yellows 271, 266, or orange yellows such as 161, 157; drops few uncolored, seen mostly in the marginal areas; odor none; conidiophores about 400 to 600 by 3 to 3.5 $\mu$ , straight, simple or somewhat branched, with all walls smooth; branches about 12 to 20 by 3 to 4 $\mu$ , two or rarely three in the group, with vesicle-like apices; metulae about 10 to 14 by 2.5 to 3 $\mu$ , in groups of 5 to 7, somewhat claviform with enlarged or vesicle-like apices; sterigmata about 8 to 9 by 2 to 2.5 $\mu$ , in verticils of 6 to 12, more or less straight; conidia 2.3 to 2.8 or even 3 $\mu$ , smooth, globose, with connective evident, not persisting in chains in fluid mounts.

Habitat: Species isolated from earth in pine woods at depth of 5 cm. in "Długa Goslina" near Poznan in Poland.

Biourge reported this culture as related to *P. commune* Thom and *P. puberulum* Bainier hence Zaleski classes it in "Subsection III Lanata." Our notes from Zaleski's culture follow: Type strain growing better at 20° than at 30°C. or over; colonies upon Czapek's solution agar slowly growing, azonate, floccose, producing a mass up to 1 mm. deep in the center and marginal area almost velvety plane, 2 to 4 mm. wide; conidial area, sage green (Ridgway XLVII and XLVI), becoming deep dark brown above in several weeks; reverse uncolored or slowly vinaceous shades; odor, none; conidiophores up to 500 $\mu$  or even 1000 or 2000 $\mu$  at margins of colonies by 2 to 3 $\mu$ , with walls smooth; penicilli variously branching producing sterigmata at several levels (stages); with lower branches

more or less diverging; metulae commonly short 10 to 12 $\mu$ ; sterigmata about 9 to 10 or even 12 by 2 $\mu$ ; conidia 2 to 3 $\mu$  in diameter.

Culture no. 5010.19 received as type from Baarn in July, 1928, complies with Zaleski's description closely enough to be accepted. In some cultures, the marginal area of the older but still growing colony have been almost arachnoid with submerged fertile hyphae bearing submerged penicilli and even chains of conidia.

*Conidiophore walls smooth: conidia elliptical.*

213. *P. aurantio-candidum* Dierckx. In Biourge Monogr. La Cellule 33: 116-119; Col. Pl. I, Cart 11, Pl. II, fig. 4, Pl. XXIII, fig. 136. 1923.

Biourge suggests as possible synonymy *P. bicolor* Fries and *P. aureum* of Van Tieghem.

Colonies on wort gelatine in the zonate group floccose (duveteuse = downy) with central area a deep floccose mass of mycelium yellow at base, white above, slowly bluish from the development of conidia, thinning toward the margin to a narrow submerged border, zonation broad and not well marked; coremial tufts are noted upon some media but not adequately described; reverse yellow to orange becoming reddish to almost bay toward the margin; substratum yellow, to reddish brown (bay); conidiophores commonly very long, commonly 3.5 $\mu$  in diameter above, 6.5 $\mu$  at base; penicillus loosely branching up to 160 $\mu$  long, figured as main branch and unequal more or less diverging branches and verticils of branches bearing metulae and sterigmata at 1 or more levels; branches 18 to 24 by 3.5 $\mu$  in twos or threes; metulae 8 to 14 by 3 $\mu$ , 2 to 4 in the verticil; sterigmata 8 to 10 by 3.5 to 4.5 $\mu$  in threes (?—C. T.); conidia elliptical 4 to 5 by 3 to 4 $\mu$ .

Biourge's no. 11 (our no. 4733.6) grew well upon liquid media, liquefied gelatin rapidly producing sulphur yellow islets floating upon the liquid, and produced conidial areas slowly upon all media we have used. When grown upon Czapek's solution agar it produced colonies broadly zonate, deeply floccose, spreading fairly widely, white with yellow or orange tinge near the substratum and thinning almost to velvety at margin tardily developing bluish gray green conidial areas passing slowly into mouse gray areas, unevenly distributed, tending to form a fairly dense zone near the margin, with tufts or clumps of penicilli or even dense central spots, scattered over a white background of aerial mycelium; reverse in unevenly distributed orange shades, persistent or some-

times passing into maroon or bay shades; penicilli up to about 40 or even 70 $\mu$ , with masses of chains up to 125 $\mu$  or longer, including branches up to 20 to 40 $\mu$ ; metulae about 12 to 16 $\mu$  and sterigmata 7 to 9 (10) $\mu$ , with elements differing in length hence producing sterigmata at different levels, with conidial chains at first parallel then forming a tangled mass in age; conidia mostly 3 to 3.5 less commonly 4 $\mu$  and globose or subglobose as given by Dierckx rather than 4 to 5 by 3 to 4 $\mu$ ; as given by Biourge although occasional longer conidia are seen. Biourge's drawings indicate conidia globose and mostly small.

There has always been a doubt in our minds about Biourge's culture. Sometimes it seems to be pure, again the characteristics of a submerged contaminant have appeared. The description given repeats in the main Biourge's characterization showing that we have approximately the same species or "compound" he had. At least once, a pure white organism with loosely radiating partly aerial partly submerged hyphae has been isolated. But its exact relation to the species remains unsettled.

215. *Penicillium biforme* Thom. Emended from U. S. Department of Agriculture Bureau of Animal Industry Bul. 118: p. 54-55, fig. 18. 1910. See fig. 45.

Cultivated in gelatin and in Czapek's solution agar white, slowly gray or greenish gray, at times with a faint fleshy or rosy tint in the marginal area in Czapek, densely floccose, with broad vegetative margin, spreading widely over the substratum; in potato agar white, then gray-green, rapidly becoming yellowish-brown, drab or almost olive, restricted in growth, aerial portion consisting of very short densely crowded conidiophores and conidial fructifications; reverse colorless; drops colorless; conidiophores 60 to 150 $\mu$  on agar or slightly longer when arising as branches from the floccose aerial mycelium growing upon gelatin; penicilli mostly once or twice alternately branched, branches convergent or divergent, each bearing a verticil of metulae crowned by verticils of sterigmata with chains of conidia, the whole 60 to 240 $\mu$ , usually 100 to 200 $\mu$  in length; sterigmata 8 to 10 or even 13 by 3 $\mu$ ; conidia elliptical or cylindrical, then globose, 4 to 4.3 $\mu$  by 3.2 to 3.5 or 4 $\mu$  in diameter, adhering in chains in fluid mounts; grows luxuriantly in fluid offering milk-sugar as source of carbon, partially and slowly liquefies gelatin, with alkaline reaction to litmus; odor, very strong, "moldy" characteristic.

Type: Thom no. 39 from French cheese.

Biourge in his Monograph (La Cellule 33: fasc. 1, pp. 194-196; Col. Pl. III, Cart. 167; Pl. V, fig. 29, 1923) discusses his no. 167 (our

no. 4733.18) which appears to be *P. biforme* as a possible synonym of *P. canescens* Sopp, although he finds the conidia too small.

Biourge adds from his wort gelatine cultures the following observations: Colonies on wort gelatine lanose, rich green, becoming more or less rosy and finally brownish; reverse orange yellow; odor strong, "intolerable," moldy; conidiophore 3.5 to 4 $\mu$  in diameter; penicillus about 35 $\mu$  long, with all walls smooth, figured as a main axis with terminal verticil of metulae and an appressed branch at the next node with accessory and smaller

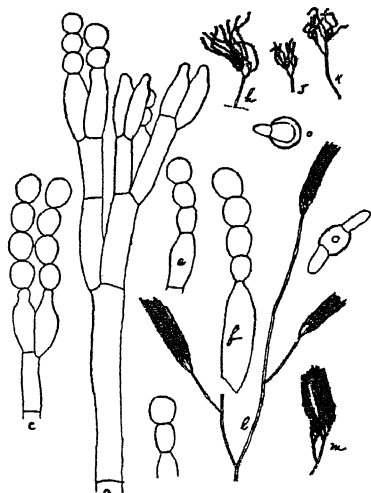


FIG. 45. *P. biforme* Thom 1910: a, c, e, f, detail of structures; h, j, k, sketch of dwarf conidiophores and penicilli from potato agar; l, m, conidiophores and penicilli from floccose colony.

conidial mass; branches 5 to 15 by 2 to 3.5 $\mu$  in pairs; metulae 8 to 16 by 2 to 3 $\mu$ , in twos or threes; sterigmata 8 to 11 by 2.5 to 3.5 $\mu$ , in verticils of 2 to 4; conidia subglobose 3.5 to 5.5 by 3.2 to 4.5 $\mu$ .

Biourge's no. 167 (our no. 4733.18) varies but slightly from Thom's no. 39 which was sent to Biourge. He comments that after strenuous attempts to purify it, he was convinced that Thom's culture was a good species whose odor was its outstanding character. Upon Czapek's solution agar the colonies were floccose piled unevenly in places as deep as 5 mm., thinning to a broad submerged margin, white then very slowly gray green with reverse not colored.



Westling (Arkiv för Botanik 11, pp. 52-53, 67-70, figs. 4 and 48, 1911) found *P. biforme* very common in Sweden, on cheese, on oak bark, on fruits, on Rhamnus, on drugs as a soil contaminant. After careful study of its contrasting forms in different media Westling agreed that the species was good.

*P. aurantio-albidum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 197-198; Col. Pl. III, Cart 47; Pl. V, fig. 28. 1923.

Colonies on wort gelatine, deeply lanose, from white to yellowish and with scanty conidia gray or on some media slightly bluish, coremia none; reverse orange yellow diffusely; conidiophore about  $3\mu$  in diameter; penicillus about  $25\mu$  long, with all walls smooth, figured as a main axis terminating in a verticil of few sterigmata with one or ? branches or metulae from the next node and frequent malformations; metulae 13 by 2 to  $3\mu$ , in pairs; sterigmata 7 to 9 by 3 to  $3.5\mu$ , in twos or threes but frequently very small, deformed, vacuolized and sterile; conidia ovate, 3.5 to 4.5 by 3 to  $3.5\mu$ .

Biourge's no. 47 (our no. 4733.4) on Czapek's solution agar produced colonies deeply floccose or lanose, 2 to 5 mm. in depth, with center deeply irregularly piled thinning to a submerged margin (1 mm. wide in cases seen), with central area greenish gray from tardy development of conidia (after several days); reverse yellowish to orange or reddish orange in age, sometimes in with deeper colors in spots, interspersed with dark gray areas; odor fairly strong, moldy; otherwise not differing from Biourge.

217. *P. lanoso-coeruleum* Thom, n. sp.

Colonies upon Czapek's solution agar floccose forming fairly closely woven felts 1 to 2 mm. deep and rather broadly spreading, usually radiately wrinkled toward the margin in the latter part of the growing period, margin broad 3 to 10 mm., becoming thin and narrow on shallow areas of the culture media but broad deep and white or slightly cream color in deep agar, traces of zonation sometimes seen toward the margin in reverse; conidial areas deep blue green (near Ridgway's bluish glaucous) in age changing through shades of olive gray to light mouse gray; reverse uncolored; odor none or indefinite; drops crystal often large and deeply imbedded in the mycelial felt and leaving large "pockets" when they dry up; conidiophores 200 to  $600\mu$  or longer by about  $3\mu$ , with walls pitted or delicately roughened (granular as seen dry under low magnifications), penicillus variously branching, usually showing a branch or branches, verticils of metulae, and sterigmata producing chains in a

more or less columnar mass, up to  $300\mu$  long at times, sometimes splitting to a series of columns or breaking to a tangled mass in age; sterigmata about  $8\mu$  long, few in the verticil; conidia mostly showing ellipticity up to 3.5 or even  $4\mu$  by  $2.5$  to  $3\mu$ , less commonly subglobose; often seen in long chains in fluid mounts.

Type no. 2543a. Name selected to indicate a floccose or lanose species in shades of blue green with the blue strongly evident although not very satisfactorily represented by Saccardo's *coeruleus*.

*Sub-section 2. Lanata-Zonata. Colonies with well marked zonation at least in the latter part of the growing period.*

218. *P. roseo-citreum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 184-186; Col. Pl. IV, Cart. 131; Pl. VII, fig. 39. 1923.

Colonies in wort gelatine, floccose felted, at first gray greenish, then dull green and finally brown with frequent overgrowth rosy; reverse at first yellow (aureus) soon red, or dark red; gelatine liquefied and reddened; drops golden yellow; odor none; conidiophore 3 to  $3.5\mu$  in diameter; penicillus  $20\mu$  without branching, 40 to  $50\mu$  with branching, with all walls smooth, figured as a main axis with terminal verticil of metulae more or less diverging, and with or without a branch from the next node bearing a secondary fruit; branches none or in verticils  $25\mu$  long; metulae 8 to 12 or even 15 by  $2.5$  to  $3\mu$ ; sterigmata 8 to 9 by  $2.5$  by  $3.5\mu$ ; conidia globose or subglobose 3 to  $4\mu$ .

Type: Biourge's no. 131 (our no. 4733.106); cultures on Czapek's solution agar, broadly hemizonate in age, floccose forming a felt, up to 3 or 4 mm. deep, becoming pale bluish green in conidial areas and later gray, producing abundant yellow drops, reverse and agar yellow to rose-salmon or reddish; conidiophores slender 2 to 2.5 or even  $3\mu$  in diameter with walls delicately pitted appearing commonly about smooth, penicillus commonly branching with branches divergent and unequal 20 to  $30\mu$  long, bearing few metulae 10 to  $20\mu$  long, also more or less diverging, swollen at the apex and bearing sterigmata about 6 to 10 by 2 to  $2.5\mu$  and conidial chains in long columns; conidia subglobose 2.5 to  $3\mu$  less commonly  $3.5\mu$ .

While the penicillus suggests the Radiata the colony characters ally this form with the zonate Lanata.

Another strain assigned here (no. 4658.36/8 from Putterill at Cape Town) produced bluish gray green colonies with floccose mycelium forming a close tough felt with a broad white margin; reverse and agar

yellow; drops yellow ; conidiophores delicately pitted, producing penicilli with branches (rami) at one or two levels (étages Biourge), frequently exceeding the main axis in length; metulae 10 to 12 $\mu$  long; sterigmata 7 to 8 by 2 $\mu$ ; conidia globose about 3 $\mu$  or with maximum 3.5 $\mu$ .

219. *P. commune* Thom. In U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, p. 56, fig. 19. 1910. See figs. 46, 47.

Colonies in gelatin or in potato or bean agar, dull green, becoming brown when old, broadly spreading, zonate, with broad white growing margin composed only of conidiophores, in the older parts becoming

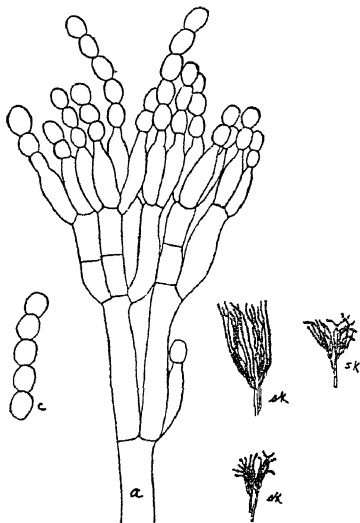


FIG. 46. *P. commune* Thom 1910: a, detail of penicillus; sk, sketches of penicilli

floccose masses of interwoven hyphae; reverse of colony and substratum never colored; conidiophores with walls rough commonly 300 $\mu$  in length, sometimes up to 700 $\mu$ ; penicilli commonly 100 to 200 $\mu$  in length, compact at base and broadening above, variously branching with branches appressed, and verticils of sterigmata 8 to 9 to 11 by 3 $\mu$ , abruptly narrowed to produce conidia; conidia cylindrical to elliptical and finally globose 3 to 4 $\mu$ , becoming 4 to 5 $\mu$  or larger in germinating smooth, green, persisting in chains in fluid mounts; colonies liquefy gelatin slowly or partially, softening rather than producing clear liquid,

alkaline in media without sugar but acid with either cane sugar or lactose, having a strong "moldy" odor.

Habitat: Common in food, dairy products, etc., Storrs, Conn.

Biourge (Monogr. La Cellule 33: fasc. 1, pp. 134-135; Col. Pl. III, Cart. 149; Pl. IV, fig. 23; 1923.) gives: Colonies on Raulin and wort gelatine, floccose, broadly and more or less definitely zonate forming a felt rather thin at the margin and up to  $500\mu$  deep in center; conidial areas at first bluish green then gray green, ultimately overgrown with white hyphae; reverse colorless or cream yellow, or with traces of rose at times; odor moldy strong; drops colorless, large; conidiophore up to  $5\mu$  in diameter, with walls smooth or slightly rough; penicillus 45 to 65 or even  $80\mu$  long, figured as a compact branching system producing sterigmata at approximately one level; branches in pairs 17 to 20 or even  $30\mu$  by 4 to  $4.6\mu$ ; metulae 14 to 20 by  $3.4\mu$ , in twos or threes; sterigmata 10 to 12 by

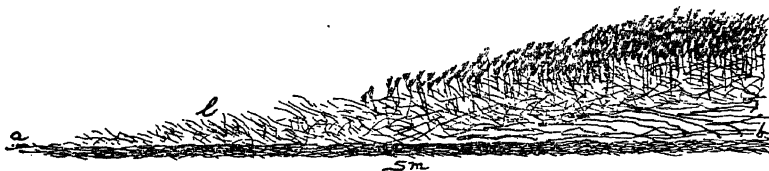


FIG. 47. *P. commune* Thom: Diagrammatic radial section of colony (magnified 25 times); *ab*, agar line; *sm*, submerged mycelium; *l*, floccose mycelium.

3 to  $3.5\mu$  in twos or threes; conidia elliptical to globose, 4 to  $6.5\mu$  by 4 to  $4.8\mu$  (in ours 3 to  $4\mu$  rarely larger).

Biourge reports this species from Brie cheese and that Dierckx had it without giving it a name; Biourge's no. 149 (our no. 4733.40, compare Thom no. 23 as described in Bul. 118). He reports his no. 41 as related and as very common at Louvain, as having a fruity, ethereal odor and to assume a rose-salmon tint on Raulin gelatine.

*P. commune*, like many other species, is inappropriately named since it is by no means the abundant or usual organism Thom supposed it to be, when he described it. It is however found often enough to say that it may be used to typify one of the diverging series of the lanate group.

220. *P. fusco-glaucum* Biourge. Monogr. La Cellule 33: pp. 128-130, Col. Pl. I, Cart. 32; Pl. II, fig. 9. 1923.

Colonies on Czapek's solution agar, with zonation fairly distinct, floccose in central area commonly  $500\mu$ , at times  $600$  to  $800\mu$  deep, with

broad white marginal area almost velvety, and central conidial area gray green to dull dusky green; reverse at times irregularly (spotted) yellow to orange, or at times with rose areas, and becoming drab in age; agar only slightly yellowed; odor slight, fragrant; conidiophore with walls rough, with apex enlarged; penicillus with walls rough,  $60\mu$  long, figured as main axis and one appressed branch often very long, unseptate or 1 to 2 septate, and bearing metulae and sterigmata at one or more levels; primary branches 20 to  $30$  by  $3.5\mu$ , appressed; metulae 15 to 18 by 3.5 to  $4\mu$ ; sterigmata 12 to 14 even to 18 by 3 to  $4\mu$ ; all parts becoming deciduous in age, and producing loose tangled chains of conidia which are quickly ripened without secondary or continued multiplication or increase in the mass of spores; conidia in our cultures about  $4\mu$ , reported by Biourge as 3.8 to 4.8 by 3 to  $4.5\mu$ , in long tangled chains.

Biourge's no. 32 (our no. 4733.64) carried as a contaminant *A. conicus* Blochwitz which may account for some discrepancies between our observations and his. In culture upon potato, Biourge reports coremiform structures at the margin of the colony. In most cultures they are reported as none or rudimentary.

Culture nos. 5034.19 and 5034.8 from Nobels Explosions Company, Ayrshire, Scotland, appears to be near to this species. As a variant of Biourge's species our notes on no. 5034.19 may be given: Colonies upon Czapek's solution agar floccose up to nearly 1 mm. deep in central area, ridged zonate in the shallower areas toward the margin, conidial areas in the outer or younger zones near *Gnaphalium* green (Ridgway XLVII) passing to olive gray or even to mouse gray in the older areas in center; reverse showing zone lines plainly, colorless to yellowish passing to pinkish or flesh color; odor none or faint; drops abundant colorless at first later pale cinnamon; conidiophores 300 to 500 or longer by 3 to  $4.5\mu$ , with walls rough, arising either from submerged or aerial hyphae; penicillus consisting of main axis and one branch about 20 to  $26\mu$  long, with metulae 12 to  $20\mu$  and sterigmata 10 to 12 or even to  $14\mu$  or with long tubes reaching 16 to  $18\mu$ , few in the verticil, produced at different levels upon the main axis and branches; with conidia in tangled chains; conidia up to 4 by  $3.5\mu$  usually showing some ellipticity but sometime almost globose, smooth, vacuolate.

In general aspect this series suggests the series of forms assigned with *P. terrestre* Jensen but lacks the funiculose hyphae characteristic of that series.

**221. *P. lanoso-griseum* Thom. n. sp.**

Colonies upon Czapek's solution agar, deeply but rather loosely and evenly floccose about 2 mm. deep and spreading widely over the substratum, zonation commonly not seen in young colonies, evident in the older colonies as ridging in the upper half millimeter scarcely going deeper, with colorless partly imbedded drops along the zone lines; colorless margin 2 to 10 mm. wide during the growing period; conidial area in dull green shades passing through gray shades toward light mouse gray in several weeks; reverse colorless; odor slight moldy penetrating; conidiophores variously produced as branches from aerial or submerged hyphae up to 1 mm. or more in length by 2 to 4 $\mu$  in diameter, with walls pitted to coarsely granular in age; penicillus more or less compact with branches short to fairly long, in a mass or column or variously splitting even becoming a tangled mass in age; metulae 12 to 20 $\mu$  long; sterigmata up to 12 by 2.5 to 3 $\mu$ ; conidia about 4 by 3 $\mu$ , commonly found in chains in mounts.

Type no. 2746.2a from leaf mold collected for us by C. J. Koning in "Spanderswoud" near Bussum, Holland, in 1913; name for a lanose species with gray conidial areas.

## CHAPTER XVII

### THE LANATA-DIVARICATA

*Section 4. Lanata-Divaricata.* This section shows the well-marked development of an apical or terminal group of unicellular branches or metulae. Such groups usually include a prolongation of the main axis, and the members of each group diverge to produce a 1-sided, or asymmetrical verticil, each member of which produces a verticil of sterigmata bearing conidial chains thus giving the appearance of a monoverticillate penicillus, that is, as if each were unmodified by the presence of the other members of the terminal group of branches. Accessory penicilli borne upon branches may appear either as simple monoverticillate forms or variously branching groups usually with the divaricate arrangement found in the terminal aggregates (fig. 48).



FIG. 48. The divaricate type of penicillus

Zaleski described certain of his species which we have placed here as "*Biverticillium subsec.*" etc. In the broad sense the term biverticillate would include every *Penicillium* with its penicillus consisting of two verticils.

As in other divisions, this section is based upon an arbitrary character, hence it is not a genetic entity. Transition from the divaricate penicillus toward the typical brush or broom with its parts aggregated to a fairly homogeneous structure occurs among the species included. Such border line species usually show other characters upon which they are placed either among the divaricate series or in one of the other sections. In such cases, the allocation has been based upon the apparent usefulness of the association to one trying to identify species.

A group of velvety species with penicilli divaricate but closely related to individual species with the more complex type of penicilli have been placed with the Velutina, already described.

*Key to species included*

- I. Colonies characterized by aerial mycelium with part or all of the conidial apparatus developing on branches from such hyphae.....II.
- I. Colonies showing trailing and anastomosing ropes or funicles of aerial hyphae in colonies otherwise floccose in general character.....See Funiculosa, Chap. XVIII.
- II. Colonies showing some shade of green, gray green or blue green with the ripening of conidia (occasional species practically colorless).....III.
- II. Colonies floccose becoming lilac and related shades variously described as rosy, violet, violaceous, etc.....*P. lilacinum* series.
  - a. Colonies approaching Saccardo's lilacinus above and below.....*P. lilacinum*, 225.
  - b. Colonies described as sordid rosy to lilac.....*P. rubellum* (Bain.) fide Biourge, 226.
  - c. Colonies "light lobelia violet".....*Spicaria violacea* Abbott, 227.
- III. Colonies with more or less green in conidial areas, (colorless variants occasional) mycelium floccose or with traces of fasciculation in some species.....III. a and b.
  - a. Penicilli with divaricate character well marked.....IV.
  - b. Penicilli tending toward compact biverticillate but with short conidial tubes and globose conidia.....XII.
- IV. Conidia globose rough, echinulate or spinulose....XV.
- IVa. Conidia globose smooth.....V.
- IVb. Conidia definitely elliptical.....XVII.
- V. Reverse definitely pigmented during the period of growth.....VIII.
- V. Reverse uncolored or color not recorded.....VI.
- VI. Conidia 2 to 3 $\mu$ .....VII.
- VII. Colonies broadly spreading loosely and evenly floccose, pale grayish green.....*P. (Spicaria) simplicissimum* (Oud), 230.





## XV. Conidia globose or subglobose rough or spinulose...XVI.

- XVIa. Colonies forming shallow felts, green to olive to brown; reverse yellow to reddish yellow; conidia 3 to 4 $\mu$  rough.....*P. albidum* Sopp, 244.  
Presumptively identified in.....No. 4942 B.
- XVIb. Colonies tending to funiculose, gray or olive gray shades with scarcely a trace of green; conidia 3 to 3.5 rough.....*P. nigricans* Bainier-Thom, 245.
- XVIc. Colonies almost velvety, dark gray (? greenish); reverse yellow; odor none or slight; conidia 3 to 3.5 $\mu$  rough.....*P. Swiecickii* Zal., 246.
- XVIId. Colonies almost velvety but with a close meshed felt; reverse yellow to orange brown; conidia about 3 $\mu$  with roughening granules more or less in bands.....*P. Janczewskii* Zal., 247.
- XVIe. Colonies forming a thin velvety felt; gray green to olive gray; reverse pale orange; conidia 2.5 to 3 $\mu$  with very delicate spinulosity.....*P. Kapuscinskii* Zal., 248.
- XVII. Colonies ochroleucus with bluish green conidial areas; conidia 3.5 to 4 by 2.7 to 3.2.....*P. Briosii* Carbone,

In these species, the aerial hyphae are not combined into ropes or fascicles (or only show traces of such ropes); most of them are floccose felted but the few reach almost a velvety appearance while showing a thin basal aerial felt. The species assigned naturally fall into several series although some forms remain as individual species showing little relationship to these natural aggregates.

*Colonies without green color but in various shades of lilac (lilacinus), floccose with elliptical to fusiform, smooth conidia, and reverse commonly in lilac shades; penicillus widely divergent in character and presenting bizarre arrangements of sterigmata as well as the divaricate type of penicillus described for the section. The P. lilacinum series, no. 225, 226, 227.*

*P. lilacinum.* Thom. U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118: 73, 74, 75, fig. 30. 1910. See figs. 49, 50.

Cultivated in pure gelatin or bean agar white, white to pale lilac, in cultures containing sugars, more or less loosely floccose with hyphae

branched, septate, ascending,  $3\mu$  in diameter, producing conidial masses upon very short branches irregularly distributed, or becoming conidiophores toward the apex; reverse of colony not discolored; conidial fructifications up to  $100\mu$  in length, consisting of solitary, sessile sterigmata, or verticils of sterigmata, or short branches bearing 1, 2, or 3 verticils of branchlets and sterigmata with long, tangled chains of conidia. Sterigmata flask-shaped, divergent at the apices, acuminate, 7 to  $10\mu$  in length; conidia elliptical, smooth, 2.5 to 3 by  $2\mu$ , thin walled, pale lilac. Colonies slowly liquefy gelatin, with strongly alkaline reaction.

Type no. 8 was received from Prof. G. F. Atkinson and C. W. Edgerton, Ithaca, N. Y.

Close relationship of this species to the common green forms is very doubtful. The chains of conidia produced break up so quickly and com-

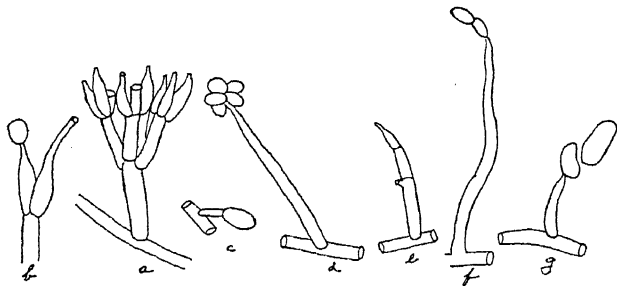


FIG. 49. *P. lilacinum* Thom: a, detail of normal penicillus; b, one normal sterigma and one growing out into a hypha in the same verticil; c, d, e, f, g, types of branching and "spore" formation found submerged in the substratum.

pletely in mounting in fluid for examination that it is often difficult to find even a single conidium attached to its sterigma.

Thom's figures 30, a, b, and c, magnify the importance of the bizarre variations presented in microscopic examination of many slides. The more usual types of branching in the soil *Penicillia* are much more common than these.

Various workers (notably Westling, 1911, p. 3) have expressed the opinion that this series should not be listed among the *Penicillia*. Abbott reported *P. lilacinum* from Louisiana as a *Penicillium* but followed it in the same manuscript by describing a nearly related organism showing more deeply violet shades as *Spicaria violacea* (our no. 4894.4). Others have expressed the same opinion as to proper generic disposition

of these forms. Without definite knowledge of their real relationship these species have a conidiophore morphology sufficiently penicillate to justify reporting them in this place until a real connection can be established.

The members of the series are constantly encountered in cultures from the soil or from soil-contaminated substances. Their conidial apparatus, although lacking green color, resembles in general morphology the series discussed here as the soil *Penicillia* sufficiently to suggest that the absence of green color is the main basis for separation. The conidia are elliptical or elliptical to fusiform and taken with the occasionally symmetrically branched penicillus might suggest assignment to the Biverticillata but their main affinities are here.

Biourge (Monogr. La Cellule 33: fasc. 1, pp. 223-224; Col. Pl. VI, Cart. 175; Pl. X, fig. 58, 1923) discussed this species but adds nothing



FIG. 50. *P. lilacinum*: diagrammatic radial section of margin of colony (magnified 50 times).

to Thom's description. No culture of it was received from him. He placed the species, however, near Bainier's *Scopulariopsis rubellus*. In this as occasionally Biourge has erred seriously since the method of spore formation differs markedly from the *Scopulariopsis* series. Moreover *P. rubellum* (no. 165) (our no. 4733.109) of Biourge is not a *Scopulariopsis*, hence not correctly identified as Bainier's species but belongs with *P. lilacinum* Thom.

Another strain (no. 4855) shows submerged clusters of spores on the tips of short branchlets. In seeking for the meaning of this observation careful following of regular conidia-producing hyphae showed several types of branchlets. Some of these are penicillate and are irregularly branching as shown in fig. 30 of Thom's Bulletin 118, still others arise as single sterigmata with single chains. Now if such single sterigmata arise below the surface of the agar, the clusters described above may be

easily developed while the shape of the individual spore is indicative of swelling toward germination.

Many cultures with the morphology and general reactions of *P. lilacinum* have been seen since the description was published in 1910. These differ in the intensity of the lilac color produced, in vigor and mass of mycelium but carry the general characters of the description. Among these were 4826c4 from Mr. Holden, Nottingham, England, Miss Dale's C14 in Ann. Mycol. 12: 43, 1914; no. 4359.84 from Brierley at Rothamsted; G. F. Atkinson's no. 22790; no. 4853 from electroplating (see Chapter IX) solutions with varying concentration of nickel salts; no. 4831 isolated from an adrenalin solution in the Bureau of Chemistry; no. 4202.19C collected by Chung in China; no. 4055.19 from New Jersey soil; no. 11719.6 from timber, Madison, Wisconsin; two from Louisiana; many others seen in bacteriological plates in the Soil Microbiological Laboratory have not been recorded.

The real relationships of this species remain for further study as well as the possibility of separating a series of related forms upon good diagnostic characters.

6. *P. (Scopulariopsis) rubellum* (Bainier) Biourge Monogr. La Cellule 33: fasc. 1, pp. 221-222; Col. Pl. VI, Cart. 165; Pl. X, fig. 56. 1923.

Synonym: *P. amethystinum* Wehmer fide Biourge.

Colonies on wort gelatine tomentose, sordid rosy to more or less lilac coremia none; reverse yellow to dark fuscous, odor weak or alliaceous, near arsenical; conidiophore about  $4\mu$  in diameter, frequently rugulose (coated with crystals ?); branches rare about  $20\mu$  long and usually diverging at a right angle; metulae 7 to 14 by 2 to  $4\mu$ , in pairs or threes; sterigmata 8 to 12 by 3 to  $4.2\mu$ ; conidia small, lemonshaped, 2.5 to 4.5 by 1.8 to  $2.5\mu$ .

Biourge's no. 174 (our no. 4733.2a) labeled *P. amethystinum* Wehmer was received instead of no. 165 which is recorded as identical. No. 4733.2a grew in Czapek's solution agar as a pale lilac colony, slowly growing, becoming a deeply floccose felt, up to 1 to 2 mm. deep, consisting of hyphae and anastomosing ropes of hyphae; reverse at margin thin, deep lilac to almost black in older areas; conidiophores without markings on walls as indefinite branches of the felted hyphae bearing a few metulae or clusters of sterigmata only with chains of conidia about 3 by  $2\mu$ .

In September, 1927, we received another culture marked no. 165 *P. rubellum* which also belongs here rather than in *Scopulariopsis*.

227. *Spicaria violacea* Abbott. Iowa State College Jour. Sci. 1 (No. 1): 26, fig. 3, 1926; see also Gilman and Abbott Iowa State College Jour. Sci. 1 (No. 3): 301, 1926.

Type received from Gilman our no. 4894.4; they describe it: Colonies on Czapek's agar floccose, spreading, surface white at first, becoming bright lavender or violet when mature; reverse colorless. Aerial mycelium abundant, consisting of a dense network of interwoven hyphae. Conidiophores arise as branches of aerial mycelium, erect, up to  $100\mu$  long, usually once or twice branched, but often short and unbranched. Conidial chains very long, up to  $700\mu$  or more in length; fructification a divergent head with both metulae and phialides or with phialides only; phialides  $6.5$  by  $2\mu$ . Conidia elliptical, smooth, hyaline,  $3$  to  $3.5\mu$  by  $2$  to  $2.5\mu$ .

From soil: United States—Iowa (3), Louisiana (2).

In our cultures this species shows most of the characters of this group with color passing to a deep violet (near Ridgway's XXVI "light lobelia violet)." Cultures retain their color for a long time and often show ascending (not erect) Isaria-like pointed hyphal columns whose significance has not been further worked out.

This species is included here without change of name because while we believed it related to those forms described as *Penicillia*, we are not satisfied that their real relationships are either in *Penicillium* or in *Spicaria*.

*Colonies mostly showing some green or greenish conidial areas (a few forms almost white); penicilli upon diverging branchlets with sterigmata and conidial chains mostly markedly divergent.*

*Conidia globose, smooth or in a few forms showing traces of granulation under the oil immersion objective in well ripened individuals. Chains of conidia parallel, diverging or tangled. Compare: Chains of conidia tending to form columns nos. 240, 241, 242.*

*Reverse of colonies uncolored.*

230. *P. simplicissimum* (Oud) Thom n. comb. *Spicaria simplicissima* Oudemans, Nederl. Kruidk. Arch. ser. 3, Vol. 2, p. 763. 1903. See also Jensen, Cornell Agr. Exp. Sta. Bul. 315, p. 493, fig. 127. See fig. 51.

Jensen's diagnosis: Colonies orbicular with alternating zones of cream yellow and dirty gray, occasionally showing a tinge of violet; vegetative hyphae creeping, very thin, septate, hyaline, dichotomously branched;

conidiophores erect,  $40\mu$  high, septate, hyaline, usually unbranched, at the most with a small side branch, on the tip constantly producing three branchlets; branchlets non-septate 8 to  $12\mu$  long, verticillate; conidia 2 to  $3\mu$  in short chains, globose.

Oudemans type not seen; description based upon strains isolated from soil on the plant-breeding plats of Cornell University, and preserved as Plant Pathology Herbarium no. 5921. After Jensen's paper was published Prof. Whetzel kindly furnished a culture from this type, our no. 2707 now lost. This grew on Czapek's solution agar as a broadly spreading deeply floccose colony, pale bluish green (Klincksieck and Valette no. 396, 397) in the conidial areas, with reverse uncolored; gelatine in water liquefied with reverse also uncolored; penicilli consisting of verticils of diverging branchlets bearing verticils of sterigmata 7 to 9

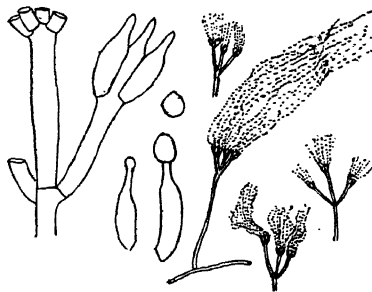


FIG. 51. *P. simplicissimum*: Detail and habit sketches

by 2 to  $3\mu$  narrowed toward sharp points at the apex; conidia  $3.5$  by  $3\mu$  to subglobose  $3.5\mu$  smooth.

Professor I. M. Lewis of Texas has recently sent in another strain with the characteristics given above (no. 5048.21a) which seems common enough to suggest that Jensen may have justly interpreted Oudemans's description hence we have introduced the change of name necessary to include this species at this point.

Colonies upon Czapek's solution agar, rather loosely floccose, spreading about 5 cm. in diameter and  $500\mu$  deep in 10 days, margin broad white; in color white to gray or in the denser conidial area perhaps a tinge of green, consisting of trailing and branching aerial hyphae, many of them very long; reverse uncolored; odor none; drops not seen; penicilli either terminal on long trailing hyphae monoverticillate, or a divaricate group of two to several metula-like branchlets, or with various mixtures

of branchlets and sterigmata in the verticil; secondary penicilli often monoverticillate; metulae or branchlets varying, mostly short; sterigmata up to 8 to  $9\mu$  long by  $2\mu$ , diverging at the tips and bearing divergent or tangled chains of conidia; conidia mostly 2 to 2.5 or  $3\mu$  in long axis, some 3.5 or occasionally about  $4\mu$ , elliptical to subglobose, showing connections in the chains.

231. *P. chrzaszczi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 464, 465, 466; Taf. 48; Zaleski no. 1645.

Colonies in neutral Raulin with 10 per cent gelatine, slowly growing, becoming 24 to 28 mm. in diameter in twelve days, and liquefying the gelatine completely, with thallus thin, irregular at margin, velvety to subfloccose, azonate, with a few (1 to 3) very conspicuous concentric wrinkles, with center either umbonate or depressed; margin (fimbria) white about 1 mm. wide during the growing period; in color at six days bluish green, C.d.C. 371, 372 becoming 367, becoming dark orange brown shades in age such as 173, 148, 147, 143; reverse azonate, irregularly concentrically wrinkled, in yellow tints such as 196, 178 D 166, 157, 171; conidiophores 300 to 600 by 2 to  $3\mu$ , straight, simple or rarely branched; penicillus 20 to  $26\mu$  long; metulae about 12 to 16 by 2.5 to  $3\mu$ , varying from 2 to 8 in the verticil, with inflated apices and either symmetrical or asymmetrical in the verticil; sterigmata fusiform about 8 to 9 by 2 to  $2.5\mu$ , commonly 8 to 10 in the verticil with short tubes; conidia 2 to 2.5 or even  $3\mu$ , smooth, subglobose to globose.

Habitat: Species isolated from earth under mixed woods with abundant herbaceous plants, in square "369 Puszcza Bialowieska" in Poland.

Zaleski notes that he placed his species at first among the *Aspergilloides* (monoverticillata) but that Biourge placed it in subsequent studies in *Biverticillium*. Admitting the culture reached Biourge in impure condition, Zaleski appears to have published the description without settling the question as to what his organism really was. Zaleski classes it "*Biverticillium* Dierckx sub-sec. 2. Concentrice-undulata." Our notes follow: Type strain growing very poorly at  $30^{\circ}\text{C}.$ , or above; colonies upon Czapek's solution agar at about  $20^{\circ}\text{C}.$ , in about ten days forming restrictedly growing, close, smooth felts, wrinkled often in quadrants, or more complexly radiately wrinkled, with white margins 1 to 1.5 mm. in width, central area greenish glaucous blue (Ridgway XLII) or related shades of bluish green; reverse not colored; odor, none; drops, not seen; conidiophores 300 to 500 by  $2.5\mu$ ; penicillus divaricate in type with a central monoverticillate continuation of the main axis with a group of



monoverticillate branches at the first septum, with branches equal or unequal in length fairly uniformly divergent, and conidial chains parallel or divergent (not adherent in columns); sterigmata 6 to  $8\mu$  long with short rather abruptly pointed tubes; conidia globose 2 to 2.5 or even to  $3\mu$ , smooth, with connective evident.

Cultures no. 5010.7 received from Baarn in July, 1928, appears to be type.

We were first inclined to put this with *P. brevi compactum* but its persistently monoverticillate penicilli, delicate hyphae in a fine felt, globose conidia 2 to 2.5, and comparison with *P. Szaferi* and *P. Bialowiezense* lead us to put it here.

As described by Zaleski *P. swiecickii* has smooth conidia and would belong here. The culture received, no. 5010.23, has echinulate conidia and must be placed with the *P. echinatum-janczewskii* lot.

*P. gratioli* Sartory, Ann. Mycol. 11: 161-165, Pl. IX. 1913.

Colonies on licorice sticks blue green (C.d.C. 353); conidiophores described as up to 5 mm. long by 8 to  $9\mu$  above and averaging 4 to  $5\mu$  in diameter and figured [from hanging drop culture apparently—C. T.] as long branching hyphae with penicillus at the apex commonly as a group of fairly equal branches or metulae diverging and each bearing a Citromyces-like fruit mass, or on short divergent branches bearing single verticils of sterigmata; sterigmata 8 to 14 by 3 to  $4\mu$  few in the verticil, diverging at the apex and bearing divergent chains of conidia; conidia elliptical to globose 2.8 by  $2\mu$ , or 2.5 to  $3.3\mu$ .

Species from the gold mines of Johannesburg. Cultures grew best at 34 to 35° but continued to grow up to 49 or 50°C. and grew well on all common media; they liquefied gelatine slowly, coagulated and peptonized milk. Sugars stand in the order of their availability for growth in this species as saccharose, maltose, levulose, lactose, galactose, inulin.

*Reverse of colonies in yellow to orange shades with or without red shades in age.*

233. The "soil" Penicillia or *P. janthinellum* series (nos. 234, to 238).

The series of "soil Penicillia" as described by Thom in Pratt's "soil fungi . . . , of Idaho" show a fairly complete gradation from simple hyphae to ropes of hyphae and all show traces of the ropy condition. It has seemed desirable to put them together at this place (Chapter XVIII) as the *P. janthinellum* series using Biourge's name for some one

of them as a collective designation for the whole lot. The most striking character of *P. janthinellum* as described is the violet shade in reverse of colonies and this characterizes a great series of strains collected from cultures on soil extract agar as used by the soil bacteriologists. Similar shades are reached through yellow and orange in Czapek's plates but more slowly (fig. 52).

Biourge in a recent note has decided that one of these cultures sent to him in December 1928 as isolated from the soil is not his idea of *P. janthinellum*. Nevertheless we believe this species to belong in the series.

Series diagnosis: Emended from Thom in Pratt in Jour. Agr. Res. (Washington) 13: 94-95, figs. 3 and 4; 1918. Colonies in Czapek's solution agar white, to gray, gray green, pale green, or pale bluish green,

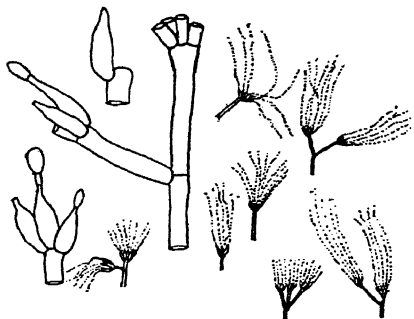


FIG. 52. *P. janthinellum* type of penicillus: Detail and habit sketches

when old becoming various shades of gray and brown, spreading slowly but broadly with usually a wide sterile margin throughout the growing period and slow development of colored fruit from the center outward, surface growth from velvety at the margin in some with center floccose to others floccose out to the very edge of the colony, with still others showing bundles or more or less definitely, ropes of hyphae, some strains zonate, reverse of colony at first colorless, in some strains remaining so, in others developing a succession of colors appearing in series so that different strains become finally yellow, orange, orange-red, rosy, or even deep red; the color mostly remains in the mycelium, hence does not discolor the agar beyond the areas of immediate contact, if at all; odor produced: none or indefinite during the growing period; often more or less offensive in age; conidiophores either rising directly from the substratum or as

branches of trailing aerial hyphae, from very short up to  $1,000\mu$  in length or longer, slender mostly, 2 to  $3\mu$  occasionally up to  $4\mu$  in diameter with walls smooth in some strains, slightly granular or roughened, in others, or showing both conditions in the same culture; conidial fructifications variously branched from a single terminal verticil of sterigmata to an asymmetrical verticil of metulae (branches bearing verticils of sterigmata) including the main stalk prolonged and a single branch or a whorl of branches; sterigmata few in each verticil, mostly slender 7 to 10 by 2 to  $2.5\mu$  narrowing to a slender tube from which the conidia are formed. Conidia at first definitely elliptical, 1 to 2 by  $2.5\mu$  becoming  $2.5\mu$  in diameter or even 3 to  $3.5\mu$  in age, continuing elliptical or becoming almost globose, either smooth or delicately roughened or both conditions in the same strain.

Habitat: Soils.

Illustrations: Strain no. 89. Colonies pale green velvety at border but more or less floccose in center with under side of mycelium rose to dark red, conidia becoming globose, 2 to  $3\mu$  in diameter; and, strain no. 2490, differing very little in structure but with reverse colors slowly yellow to orange.

This description was based upon a series of forms contributed by Zundel in 1916 (our no. 4157, sub-numbers 401, 452, 597, 598, 601, 604, 606, 675, 756), but has been supplemented by many cultures isolated by ourselves and others in succeeding years. The species included here are these described by various authors which seem to fall in this group.

A colony of one of these forms no. 4141 grown in Czapek's solution agar showed a range of color in the substratum from a rich yellow with a pH of 5.3 (quinhydrone electrodes) to a deep dark red with a pH of 7.3.

Some of the forms as encountered in culture seem to be well fixed and describable, others either to be unstable or to respond to slight environmental changes to such degree as to baffle attempts to describe them in concrete terms. While they seem to fall naturally into one series with smooth walled conidia, and another with rough conidia, an occasional culture has seemed to show both types of conidia at times. Since the describers have given the markings of the spores when observed, the described species may be separated on that basis but there is a doubt if the distinction is really a stable one.

Sopp seems to have had several closely related forms with rough spores in his collection when he described and figured *P. albidum*, *P. acidoferum*, *P. deformans*, then figured *P. lemoni* and *P. glauco-griseum* with rough walls as their chief separating character. Apparently these

belong near *P. echinatum* Dale (*P. nigricans* Bainier) which we have actually handled.

At best, identification of a particular culture with some definite one of these species from the available data may be unsatisfactory if not impossible since it may either fit one of them approximately or equally likely diverge in some of its characters from all of them.

234. *P. janthinellum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 258–260; Col Pl. VII, Cart. 37; Pl. XII, fig. 70. 1923.

Colonies on wort gelatine bluish green, gray green or bright green, azonate, with surface growth consisting of networks of hyphae and ropes of hyphae, tardily becoming reddish (“rubicante”); coremia none; reverse yellow to ochraceous, odor weak, conidiophores 30 to 40 by  $2\mu$  arising from creeping hyphae or ropes of hyphae, with all walls smooth; penicillus a single one-sided verticil of metulae with occasionally one branch from a lower node, hence short about  $15\mu$  long or 30 to  $50\mu$  when branched; metulae 7 to 10 by 1.5 to  $2\mu$  mostly in threes; sterigmata 5.5 to 9 by 1.5 to  $2\mu$ , in pairs or threes, apparently the *Eupenicillium* type; conidia globose 2.4 to  $3\mu$ .

Biourge’s type no. 37 was not received. See the preceding paragraphs and the following characterization of a strain of this series which was intensively studied by Dr. S. A. Waksman in New Jersey.

No. 4789, isolated by Waksman from New Jersey soil shows structures close to *P. janthinellum*: Colonies on Czapek’s solution agar containing 3 per cent sucrose, forming a dense tough felt of fine hyphae with margin of growing colony broad white and an intramarginal zone of yellow, fading to colorless in center, with surface unevenly tufted and with anastomosing ropes of hyphae; conidial areas slowly developing, pale gray to glaucous gray (Ridgway XLVIII.); drops colorless to brownish visible in zones in the growing colony; reverse of culture pale yellow to reddish orange and finally brown, in milk cultures golden yellow; conidial apparatus terminal or mostly on short branches of trailing hyphae, consisting of simple verticils of sterigmata (“*Citromyces*”-like) or of verticils of unequal metulae, or of verticils containing both metulae and sterigmata; metulae mostly 2 to  $3\mu$  in diameter, 8 to  $20\mu$  in length with apex vesiculose; sterigmata diverging, enlarged at base then tapering to a fairly long narrow beak-like conidia-bearing tip; conidia at first elliptical-fusiform to obpyriform, becoming nearly globose but somewhat apiculate about  $3\mu$  in long axis, thin walled smooth in divergent or tangled chains not in columns.

235. *P. Rivolii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 471, 472, 473; Taf. 50; Zaleski no. 1537.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 27 to 30 mm. in diameter in twelve days, liquefying the gelatine quickly and completely, surface growth velvety, plane and zonate only in the outer area with radiate wrinkles 3 to 5 mm. wide and 2 to 3 mm. high running toward an elevated center more or less overgrown with secondary loose mycelium; margin light rosy 2 to 3 mm. wide formed of dense mycelium; in color conidial areas in green shades in age such as 342, 343, 347, 338, 342, becoming orange brown shades in age such as 159, 153; reverse and liquefied gelatine in orange yellow shades such as 141, 137, 171, 136, 137, 134; odor none; conidiophores variable 100 to 600 $\mu$  by 2.5 to 3.5 $\mu$ , simple or sparingly branched with walls smooth; penicilli figured as monoverticillate, divaricately branched, or as having metulae or branches in the same verticil as sterigmata; branches rare distant and unequal, perhaps 20 to 35 $\mu$  by 3 to 4 $\mu$ ; metulae when present varying greatly in length and arrangement 10 to 20 by 3 to 4 $\mu$ , with apices enlarged, in groups of 1 or 2 up to 7 or even 9; sterigmata about 9 to 10 by 2.2 by 2.5 $\mu$  often very variable in size in the same group, in verticils of about 6 to 12, with rather long tubes; conidia about 2.3 to 2.8 $\mu$ , smooth globose, with connective evident. Habitat: Species isolated from earth under pine woods in square "369" of the forest "Puszcza Bialowieska" in Poland.

Zaleski named the species after the late Professor Rivoli of Posnan, Poland. He reported it to Biourge as belonging to the *Aspergilloides* and this conclusion was confirmed by Biourge but later rejected by Zaleski from further studies of his own from which he classes it in "*Biverticillium* subsec. 3. Radiate-undulata." Our notes follow: Type strain growing better about 20° than about 30°C.; colonies upon Czapek's solution agar at 20°C., broadly spreading, more or less elevated in center and radiately wrinkled zonate in the outer areas in old cultures with an overgrowth of gray hyphae and ropes of hyphae in age, with conidiophores arising partly from the substratum and partly from loose network of trailing aerial hyphae with conidial areas bluish green to gray green becoming deep olive gray in age (one month); reverse yellowish to orange yellow or tawny shades in age (Ridgway XXIX.); conidiophores arising from substratum and 100 to 200 $\mu$  long or as very short branches of trailing hyphae, ascending rather than erect; penicillus either a single verticil of a few diverging sterigmata, or with one or more diverging branches sometimes with secondary branching, and with chains of

conidia diverging or tangled; sterigmata commonly about  $7\mu$  long in typical verticils, but partly up to 10 to  $14\mu$  in variously mixed groups of sterigmata and branches of varying length; conidia 2 to  $2.5\mu$  in diameter, subglobose with connective evident.

Cultures no. 5010.20 received as type from Baarn in July, 1928, complies with Zaleski's description well enough to be accepted.

236. *P. cavum* Sopp. Monogr., pp. 192-194; Taf. XXIII, fig. 36. 1912.

Colonies on meat-peptone-sugar-gelatine, white with a trace of yellowish, with the development of conidia clear blue-green to olive-green, dark olive green and finally brown, with mycelium rather thin, flat or slightly wrinkled or buckled at the center; hyphae delicate; reverse at first colorless, then with a reddish tinge and finally brown, gelatine slowly liquefied, and colored red to coffee brown; odor objectionable, of the cow-stall; conidiophores much branched, inconspicuous on the mycelium, with short branches partly *Citromyces*-like, part *Aspergillopsis*-like, bearing verticils of 6 to  $8\mu$ , short swollen metulae bearing verticils of sterigmata and spore chains, each suggestive of *Citromyces*; conidia globose, smooth 3 to  $4\mu$ , produced in great abundance; perithecia not found.

Species found upon a "beerquirl" in a cellar in Ostland. Cultures grew best at  $20^{\circ}$ , with minimum at  $3^{\circ}$  and maximum at  $28^{\circ}\text{C.}$ , and grew well on Sopp's usual media.

237. *P. guttulosum* Gilman and Abbott. Iowa State College, Jour. Sci. 1: 298, fig. 33. 1927.

Type received from Gilman, our no. 4894.16: Colonies in Czapek's solution agar, floccose 1 to 2 mm. deep enmeshing many drops yellow through amber to deep purple red, with the mycelial mass buff or yellowish to lilac in the central areas; reverse orange; penicilli borne upon very short branches or terminal on aerial hyphae, as single verticils of sterigmata, as a primary terminal verticil with main axis prolonged to produce a secondary verticil, or as a terminal cluster or verticil of branches bearing monoverticillate penicilli or occasionally one of them again branched; sterigmata 8 to  $12\mu$  by about  $2\mu$  (occasional very short forms are found), tapering to a rather broad conidial tube, closely aggregated at base and diverging at the tips or variously prolonged into slender hyphae; conidia 2 to  $3\mu$  becoming globose and dark when ripe.

The above description is redrawn from our own cultures. A culture with the same characters was isolated by Waksman (our no. 4136T5) from New Jersey soil.

3. *P. glauco-roseum* Demelius. Verhandl. Zool. Bot. Gesellsch. Wien 72: 72, fig. 3. (1922) 1923.

Colonies in prune gelatine floccose, gray-green (C.d.C. 347 to 78e), mycelial hyphae, conidiophores, branches, and metulae showing vesicular cells filled with rosy crystals and granules; conidiophores 36 to 240 $\mu$  by 2 to 6 $\mu$ , bearing a single verticil of sterigmata or a penicillate branching system at the apex, sometimes metulae arising from a red vesicle and showing irregular or antler-like branching with conidium formation suppressed; metulae 14.4 by 6 $\mu$ ; sterigmata 8.4 to 9.6 by 2 $\mu$ ; conidia globose 2.5 to 3.6 $\mu$ , smooth.

Species found in preserved Rumex and tomatoes, with calcium oxalate in colorless granules; upon paradise-apples with *P. atramentosum* Thom., and a year later on cooked elder berries, but with conidiophores mostly normal, mycelium yellower, and the yellowish red granules mostly outside the hyphae.

Demelius obtained her material from highly acid (oxalic) preserves, hence the unique structures described may be interpreted as pathological. The relationship of this species is doubtful but it appears to fit better with the series of species common in soil studies than with other groups.

9. *P. Soppi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 476, 477; Taf. 51; Zaleski no. 1437.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, quickly growing becoming 40 to 45 mm. in diameter in twelve days. Liquefying gelatine slowly but completely, subfloccose, plane and indistinctly zonate only in the outer band or zone of 2 to 5 mm., for the most part wrinkled in the center cerebriform becoming radiate outwardly toward the margin, margin 1 to 2 mm. wide; in color white then showing a conidial band near the margin yellowish green no. 271, 262, to green 347, to dark shades of orange brown or gray such as no. 147, 148; reverse in orange yellow shades such as no. 191, 157; odor weak, agreeable; conidiophores 500 to 800 or 1000 $\mu$  by 2.5 to 3.5 $\mu$  with apices enlarged and with all walls smooth; penicilli commonly 18 to 24 $\mu$  long; branches very rare; metulae enlarging from base to apex, about 10 to 14 by 2 to 4, in groups of 5 to 7; sterigmata about 9 to 10 by 2 to 2.5 $\mu$ , in verticils of 6 to 10; conidia about 2.2 to 2.5 $\mu$ , smooth, mostly subglobose, occasionally globose; Habitat: Species isolated from the earth under pure pine woods in square "652" of the forest "Puszcza Bialowiecka" in Poland.

Zaleski places it in "Biverticillium Dierckx subsec. 4. Cerebriformi-terradiate-undulata. Our notes follow: Type strain growing well at

both 20°C., and 30°C., and slightly better on wort than Czapek agar. Colonies upon Czapek's solution agar about 20°C., slowly growing, floccose with loose cottony hyphae rather than a close felted appearance, commonly wrinkled radiately, often in quadrants, with a white margin 1 to 2 mm. wide, with conidial areas greenish gray; reverse yellowish toward flesh color or finally fawn; penicilli partly as single verticils of sterigmata, partly variously branching as a terminal divergent group or 2 to several monoverticillate branches, often with secondary unequal branches farther back upon the conidiophore; sterigmata 7 to 10 by  $2\mu$  in compact groups; conidia 2.5 to  $3\mu$  smooth or occasionally showing traces of granulation.

Culture no. 5010.34 received as type from Baarn in July, 1928, gives observations which lead us to place it among a different lot of species than Zaleski.

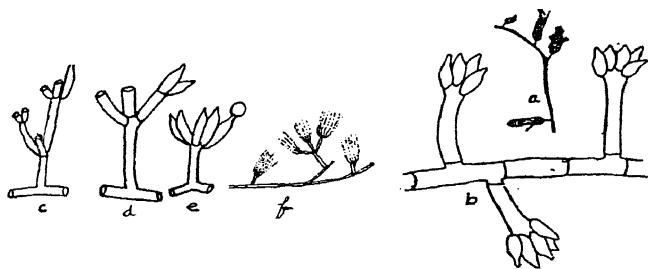


FIG. 54. *P. gilmanii* Thom: a and b, Gilman and Abbott's detail and habit sketch; c, d, e, our detail sketch; f, habit sketch.

1. *P. Gilmanii* Thom. n. sp.

Synonym: *P. cinerascens* Biourge in Gilman and Abbott Iowa State Coll. Jour. Sci. 1 (3) p. 297, Fig. 31. 1927. Certainly not Biourge's no. 90, received from him in September 1927, see species description no. 61a, Chapter XII.

Our transfer of Gilman and Abbott's organism no. 4894.12 when grown upon Czapek's solution agar, failed to show *ropiness of hyphae* as indicated in their description, at least failed to be pronouncedly so. Our diagnosis follows (compare fig. 54): Colonies upon Czapek's solution agar deeply floccose, becoming 3 to 4 cm. in diameter, 2 to 3 mm. deep in fourteen days, enmeshing large numbers of fairly large drops (colorless), aerial growth to the very edge; margin fairly deep, white, then gray (Hathi gray, Ridgway LII) with outer areas full of drops and deeper than the



darker, grayer interior; reverse with a pale yellow zone near margin; conidiophores produced as very short branches (about  $20\mu$  long) scattered along floccose aerial hyphae, singly, opposite, scattered or rarely with penicillus terminal to main hyphae; penicillus unbranched or with diverging groups of branches or metulae producing a mass of conidial chains in loosely columnar form often appearing almost solid tending to produce a column for each verticil of sterigmata; sterigmata 5 to 6 to  $7\mu$  long, from 2 to 3 in the verticil to densely crowded and fairly diverging; conidia globose or nearly so 2 to  $2.5\mu$  in diameter, in parallel to tangled chains.

Type no. 4894.12 from Iowa soil.

*Chains of conidia more or less adherent into columns; nos. 240, 241, 242.*

240. *P. glauco-griseum* Sopp. Monogr., pp. 189-190; Taf. XXI, fig. 147; Taf. XXIII, fig. 35. 1912.

Colonies on meat-peptone-sugar-gelatine, at first with mycelium white, thin, wrinkled or buckled, becoming pale blue-green to blue-green with the development of conidia; hyphae very delicate; reverse white with a yellowish tinge; odor indefinite; conidiophores enlarged upward, very coarse, rough, long, from Sopp's figures producing either a single verticil or superposed verticils of metulae variously clavate or vesiculose, either bearing verticils of sterigmata directly or partly again proliferated to bear secondary metulae; sterigmata not described; conidia elliptical to globose 3 to  $3.5\mu$  in long axis; perithecia not found.

Species found in earth. Cultures grew best at  $20^{\circ}$ , with minimum at  $3^{\circ}$  and maximum at  $37^{\circ}\text{C}$ . and grew well upon the media regularly used by Sopp. Conidia remained viable more than three years. The roughness of stalk shown by Sopp may be only the deposit of granules on its surface.

241. *P. Jenseni* Zaleski. In Bul. Acad. Palonaise Sci.: Math et Nat. Ser. B, 1927, pp. 494, 495; Taf. 57; Zaleski no. 1425.

Colonies in neutral Raulin with 10 per cent gelatin in petri dishes, fairly rapidly growing, becoming 35 to 40 mm. in diameter in twelve days, liquefying the gelatine quickly and completely, velvety and zonate at the marginal areas only, variously overgrown with trailing hyphae, and thrown into wrinkles at the older areas, with center raised, umbilicate; white margin (fimbria) 4 to 6 mm. wide, very conspicuous; in color tardily (in about ten days); showing conidial areas in green shades about

C.d.C. 322 quickly passing into orange brown shades such as 138, 139, 143; reverse and liquefied gelatine in pale yellow to orange shades such as 203A, 178D, 171, 166, 161; odor none; conidiophores 10, 20 to 300 or 500 $\mu$ , by 2 to 3 $\mu$ , commonly arising from trailing hyphae as very short to long branches, mostly flexuous, frequently themselves branched, usually inflated at the apex; penicilli 6 to 10 when simple, 15 to 25 $\mu$  long when branched, with all walls smooth; metulae 8, 10 to 15 by 2.2 to 3 $\mu$ , commonly enlarging from base to apex and inflated at the apex, and usually unequal in length and asymmetrically arranged in groups of 2, 3 to 6 or 7; sterigmata about 7 to 8 by 2 to 2.5 $\mu$ , in verticils of 3, 5 to 12 or 15, with short tubes, varying considerably in number and in size and shape according to the place occupied in the verticil; conidia 2 to 2.5 $\mu$  (or 2.8), smooth, globose, showing connectives.

Habitat: Species isolated from soil in the forest Bialowiesensis ("Puszcza Bialowieska") in Poland.

Zaleski puts it among the "Aspergilloides Wehmer Dierckx Series 1: *A. Corylophilum*." Our notes follow: Type strain growing better at 30°C. than at 20°C.; colonies upon Czapek's solution agar, at 30°C., closely felted, restrictedly growing, more or less buckled and radiately wrinkled, with margin plane, almost velvety; olive gray (Ridgway LI.), near C.d.C. 273; reverse, colorless to pale rosy (light vinaceous cinnamon of Ridgway XXIX.) suggesting the "peach" color of Bainier and Sartory's *Citromyces ramosus*; odor, indefinite; conidiophores appearing as long trailing hyphae about 2 $\mu$  in diameter with terminal penicilli or as short branches less than 100 $\mu$  long upon fertile hyphae, penicillus either monoverticillate or with various branching at the first septum or farther back on the main axis, each verticil of sterigmata producing a closely parallel almost solid column of conidial chains, up to 200 by 10 to 20 $\mu$ ; branches when clustered at the first septum 8 to 10 by 2 to 4 $\mu$ , enlarging upward, larger when borne at lower septa; sterigmata about 8 by 2 to 2.5 $\mu$ , with short tubes; conidia about 2.5 $\mu$  in diameter with connectives evident.

Culture no. 5010.10 received from Baarn in July, 1928, appears to be type.

242. *P. canescens* Sopp. Monogr., pp. 181-182, Taf. XIX, fig. 136; Taf. XXIII, fig. 28. 1912. See fig. 55.

Colonies on meat-peptone-sugar-gelatine white, fibrous ("filzig" cf. "woolly" in *P. camemberti* with which it is compared), becoming very slightly bluish or greenish with the tardily developing conidia; hyphae

delicate, fine, much finer than in *P. camemberti*; reverse white, in age brownish; substratum becoming purple brown; odor obnoxious, suggesting mouse urine; conidiophores short very slender, bearing few branches and sterigmata figured as irregularly viberticillate and more or less divergent, the branches in the verticil often differing in length, and with vesicle-like apex; sterigmata few in the verticil, pointed, with the chains in each group figured as forming a column; conidia bluish, smooth, globose, 2.5 to 3 $\mu$ , in chains figured as closely appressed (? adherent ?); perithecia not found.

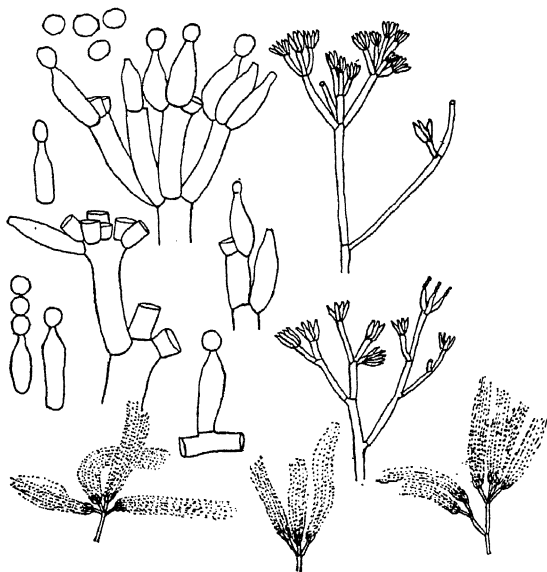


FIG. 55. *P. canescens* Sopp, interpreted Thom: Detail and habit sketches

Species obtained from earth in western Norway. Cultures grew best at 20°, with minimum at 5° and maximum at 40°C., grew well in Sopp's gelatine and agar, in milk with the development of a yellow brown mycelium and a rancid odor, well also in broth, wort, potato and rice. Conidia survived in culture more than three years.

The following characterization of a culture received from Miss Dale in England in 1912 seems to satisfy Sopp's description of *P. canescens* (no. 2654): Colonies on Czapek's solution agar with cane sugar, deeply

floccose white to yellowish then blue green no. 396-7 in Klincksieck and Vallette's Code, later grey green, slowly spreading, margin becoming more or less zonate, and thin in old cultures; reverse yellow to orange, later becoming dark reddish brown; agar less colored; conidial fructifications borne either upon special branches arising from the agar and varying greatly in length or upon short branches of aerial hyphae, consisting of variously arranged divergent branches, 1, 2, 3-verticillate; ultimate branchlets (metulae) bearing sterigmata unequal 7 to 10 by  $4\mu$ , producing chains of conidia which form more or less divergent columns in old cultures; conidia, elliptical to globose 2 to  $3\mu$  in diameter or 2 to 2.5 by  $3\mu$ , sometimes very delicately roughened, in chains which break up when mounted in alcohol.

Gelatine without sugar is rapidly liquefied so that gray green colonies orange to red in reverse float in pools of alkaline, colorless liquid with odor produced by *P. stoloniferum* which this species resembles in gelatine cultures. Crystals appear in gelatin as large concretions.

Obtained from soil in England by Miss Dale in 1912.

3. *P. matris-meae* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 477, 478, 479; Taf. 45 and 52, Zaleski no. 1433b.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, fairly quickly growing becoming 38 to 40 mm. in diameter in twelve days, liquefying the gelatine fairly quickly and completely, with surface velvety, zonate in the marginal areas undulate or wrinkled giving a cerebriform effect in the central area and becoming radiate toward the margin; margin white, fairly wide, 2 to 3 mm. in the growing colony; in color conidial areas blue green shades such as 371, 366, when young, later green 347, 341, 342, 348, 321, with green colors fading to leave the old colonies in pale orange shades such as 103B, 121; reverse in orange-yellow shades such as 171, 157, 147; odor faint, agreeable suggesting *Viola odorata*; conidiophores 300 to 600 by 3 to  $4\mu$ , straight or slightly flexuous, simple or sparingly branched, with apex dilated into a vesicle; penicilli 20 to 24 or even  $28\mu$  long, all walls smooth; metulae 11 to 13 or even to 18 by 2.5 to  $3.5\mu$ , in groups of 4 to 7, mostly enlarged upward; sterigmata about 9 to 10 by 2 to  $2.5\mu$ , or smaller, most commonly in verticils of 6 to 8; conidia about 2.5 to  $3\mu$ , smooth subglobose.

Habitat: Species isolated from earth under pine trees in square "652" of the forest "Puszcza Bialowieska" in Poland.

Zaleski notes that Biourge regarded this species as new. He places

it in "Biverticillium Dierckx Subsec. 4. Cerebriformiter-radiata-undulata." Our notes follow: Type strain growing about equally well at 30° and 20°C.; colonies upon Czapek's solution agar at 20°C., 25 to 30 mm. in diameter at fourteen days. More or less faintly zonate in the marginal areas, nearly plane or at times with several radiate wrinkles, consisting of a floccose felt bearing the conidiophores as branches, somewhat elevated in center, in gray green shades such as light celandine green or Gnaphalium green (Ridgway XLVII.); with white margin 2 to 3 mm. wide uneven fimbriate; reverse gray or cream to yellow reddish brown, with a deep brown area in center and colorless marginal areas; odor, none; drops, not seen; conidiophores 100 to 200 $\mu$  long, as vertical branches of aerial hyphae; penicilli either symmetrically biverticillate or variously reduced by absence of units or uneven development in verticils; sterigmata about 10 $\mu$  long, lanceolate; conidia mostly subglobose 2.5 to 3 as given by Zaleski but showing also elliptical conidia up to 3.5 or even 4 $\mu$  in long axis; persisting in chains in mounts.

Culture no. 5010.14 received from Baarn in July, 1928, agrees with Zaleski's description closely enough to be regarded as type.

*Conidia globose, rough or spinulose.*

244-248. The *P. nigricans-Janczewskii* series: Dale in 1912 sent us the organism later published as *P. echinatum* Dale (*P. nigricans* Bainier).

We isolated many related strains from American soil with tentative descriptions under various numbers. Later we obtained Bainier's culture as a nomen nudum—a tube with a name on it and containing a strain essentially identical. In 1924, we received Zaleski's collection from Baarn and found three more of them. *P. albidum* Sopp was added from Sopp's description. There is enough individuality among strains to separate them when carried in parallel culture; there is so much in common among the lot that they must be considered a genetic series.

*P. albidum* Sopp. Monogr., pp. 186-187, Taf. XXI, fig. 144; Taf. XXIII, fig. 33. 1912.

Colonies on meat-peptone-sugar-gelatine, at first a white, thin mycelial growth, with conidium formation clear green, later olive green, gray and finally brown, and commonly soon overgrown with mycelium, with surface growth spreading broadly, uneven, fibrous, rough, consisting of irregularly branching and trailing hyphae; reverse clear reddish

yellow, or on some media yellow; odor suggestive of paraffin; conidiophores (from Sopp's figures) arising as branches from trailing or ascending hyphae, from very short to fairly long, with walls figured as smooth, enlarging to vesicle-like apices and producing either *Citromyces*-like verticils of sterigmata or divergent clavate uneven metulae each bearing a *Citromyces*-like cluster of sterigmata; sterigmata not described and not sharply figured; conidia globose, rough, 3 to 4 $\mu$ : perithecia not found.

Species found in earth in West Norway. Cultures grew best at 20°, with minimum at 3° and maximum at 33°C., and grew well in the media tested. Conidia remained viable more than three years.

Culture no. 4894.10 received as *P. albidum* Sopp from Gilman and Abbott (cit. p. 295) proved upon transfer to belong to the *Aspergillus nidulans* group. The figure given by Sopp is that of a divaricate *Penicillium* rather than an *Aspergillus*. The following description for a culture received from Dr. McClennon at Melbourne, Australia (no. 4942B), satisfies the requirements of Sopp's species as we read them much more closely than does *A. nidulans*.

Colonies on Czapek's solution agar plane forming a closely floccose mat of fine hyphae spreading slowly over the agar with a white margin 5 to 10 mm. wide, bluish gray (Ridgway's Hathi gray LII.), shading in center to pallid purplish gray (Ridgway, LIII.) and becoming brown in age; reverse purplish vinaceous to purplish drab shades becoming brown in age; fertile branches lateral on aerial hyphae which also bear terminal penicilli consisting of metula-like groups of diverging branches and verticils of sterigmata 8 to 10 $\mu$  long, bearing conidia fusiform to globose about 3 to 3.5 $\mu$  in long axis, smooth under low magnifications or slightly roughened or pitted when highly magnified, becoming brown in age.

*P. nigricans* Bainier published here.

Synonym: *P. echinatum* Dale, Biourge, Monogr. La Cellule 33: fasc. 1, p. 278; Col. Pl. XI, Cart. 354; Pl. XVIII, fig. 104; 1923.

Discussed without name as *C*<sub>3</sub> by Dale, Elizabeth (Ann. Mycol. 12: 1, p. 42, Pl. III, figs. 51, 52, 1914; and named by her (ibid., 24: 1/2 p. 137, 1926). See fig. 56.

Colonies upon Czapek's solution agar forming a close textured fairly deep felt of fine or delicate trailing hyphae and occasionally but not regularly bundles or ropes of hyphae, plane or increasingly wrinkled at higher temperatures toward 30°C., azonate at first then more or less zonate at margin; conidial areas in various shades of gray, steel gray,

dark olive gray, Hathi gray (Ridgway LI and LII), without or with only traces of greenish, to mouse gray in age; reverse yellow to deep orange to deep ferruginous shades, at various ages and under varying conditions; odor strong, suggesting certain species of *Actinomyces*; drops abundant colorless or slightly yellowish; conidia bearing hyphae variously short branches of aerial hyphae, or whole trailing hyphae showing thickened walls and bearing short branches with penicilli; or as separate conidiophores arising directly from submerged hyphae in marginal areas; penicilli terminal on trailing hyphae or on short branches perhaps  $50\mu$  long, consisting of variously diverging branchlets bearing sterigmata few to many and chains of conidia parallel to more or less divergent or tangled in age; conidia 3 to  $3.5\mu$  in diameter, globose, echinate or spiny.

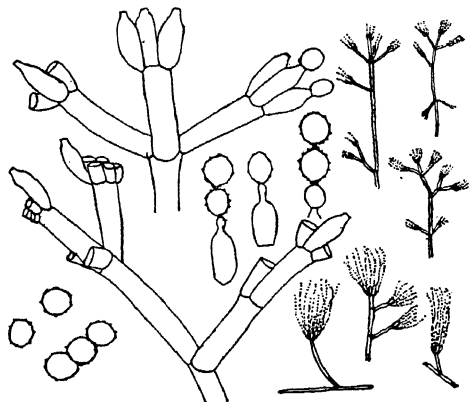


FIG. 56. *P. nigricans* Bainier: Detail and habit sketches

The characterization given was drawn from our own cultures of no. 4876.14 (type?) as received from Westerdijk at Baarn labeled *P. echinatum* Dale. Biourge's no. 354 (type?) was not received by us, but Miss Dale's C3 (Thom 2695) identical with her E3, and which appears to be referred to as type is still in our collection. It is identical with our 76 received from Prof. G. F. Atkinson at Ithaca, N. Y., in 1912, also identical with 4640.448 which came to us from the Bainier collection in Paris bearing the label *P. nigricans*.

The use of the name *P. echinatum* by Rivolta in 1873, invalidates Miss Dale's use of it in 1926 for this organism. Meanwhile we have had several strains of it including one from the Bainier collection labeled

*P. nigricans* (our no. 4640.448) but not described. Since this culture has doubtless been passed to other laboratories under the name, we are publishing it here.

Miss Dale sent her original isolations to us and published part of our notes but without name; she appears to have sent the type strain later to Westerdijk and to Biourge with the name attached. Biourge's monograph is then the first publication of the name and is followed by Miss Dale's note in 1926. We have handled in comparative culture a whole series of strains in which there are only minor variations and which may therefore be regarded reasonably as one species.

The name *P. echinulatum* Dale appears in Biourge's "List Onomastique" but not in Dale's papers. It is not explained.

Gilman and Abbott (p. 293) assign the name *P. echinatum* Dale to Thom's description of the green members of the subsection *Divaricata* in the series published by Pratt as the "Soil *Penicillia*." That description was purposely made broad enough to cover a whole series of forms which we did not wish to attempt to describe as individual species but did not include the rough-spored forms of this series.

246. *P. Swiecickii* Zaleski. In *Bul. Acad. Polonaise Sci.: Math. et Nar. Ser. B*, 1927, pp. 474, 475, 476; Taf. 51; Zaleski no. 1436.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 28 to 33 mm. in diameter in 12 days, liquefying the gelatine quickly and completely, characteristically wrinkled cerebriform in very center of the mass, thin, becoming radiate; overgrowth of secondary mycelium in the central area slight, hirsute; margin velvety azonate thin showing red from the reverse color; in color conidial areas at first blue green 371, 372 becoming dark shades of orange brown such as 138, 139, 134 in about two weeks; reverse at first in yellow to orange shades such as 181, 156, 126, 106, 101; drops uncolored, small, few; odor none; conidiophores 300, 400 to 600, even 800 $\mu$  by 3 to 3.5 $\mu$ , straight, simple or rarely branched; penicilli when unbranched 20 to 28 $\mu$ , when branched 40 to 56 $\mu$  long, presenting anomalies of mixed verticils of metulae and sterigmata, and of variations in units in the verticil; branches varying from 18 to 35 by 2.5 to 3 $\mu$ , common, enlarged at apex, usually unequal in length and asymmetrically placed one or two at the node; metulae varying in length in the penicillus from about 10 to 20 $\mu$  by 2.5 to 3.5 $\mu$ , in groups of about 3 to 6 in more or less symmetrical verticils; sterigmata varying greatly in size but mostly about 8 to 9.5 by 2.2 to 2.8 $\mu$ , in verticils of about 6 to 10; conidia about 2.3 to 2.8 $\mu$ ,



smooth, subglobose to globose with rather long tubes, and with connectives evident.

Habitat: Species isolated from earth under pine woods in square "652" in the forest "Puszcza Bialowieska" in Poland.

Zaleski reports that after thorough investigation Biourge decided this species to belong with *P. rugulosum*. This placing is clearly impossible if the description and especially the measurements given by Zaleski are correct. The conidia of the *P. rugulosum* series are elliptical and definitely roughened. Nevertheless Zaleski places it in "Biverticillium Dierckx subsec. 4. Cerebriformiter-radiate undulata." Our notes follow: Type strain growing poorly at 30°, quite well at 20°C.; colonies upon Czapek's solution agar, forming a felt of very fine hyphae, slowly growing azonate or faintly zonate in the outer areas in age, velvety or nearly so with gray fibrous overgrowth in age, plane or slightly radiately wrinkled, thin, about 200 $\mu$  deep, with white margin rather narrow and conidial areas dark gray green (near "Storm gray" Ridgway LII); reverse yellow, though orange to reddish orange; drops, numerous, small, well distributed; odor, slight, difficult to characterize; conidiophores 100 to 300 by 2 to 3 $\mu$ , with walls smooth; penicillus an asymmetrical verticil of 2, 3, 4 or more divergent branches 15 to 20 $\mu$  long, carrying verticils of sterigmata and diverging coiling chains of conidia; sterigmata 10 to 13 $\mu$  long with long tapering tubes; conidia 3 to 3.5 $\mu$  spinulose (given by Zaleski as 2.3 to 2.8 $\mu$  smooth).

Culture no. 5010.23 received as type from Baarn in July, 1928, differs from Zaleski's description in having conidia delicately spinulose and larger. If correctly described by Zaleski the species would be close to *P. chrzasci* Zal. but if this is really the type culture it belongs near *P. echinatum* and *P. Janczewskii*.

247. *P. Janczewskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 488, 489, 490; Taf. 55; Zaleski no. 1318.

Colonies in neutral Raulin with 10 per cent gelatine, in petri dishes, rather slowly growing, becoming 23 to 25 mm. in diameter in twelve days, liquefying the gelatine quickly and completely, velvety, zonate in the broad outer plane area; areas within depressed with narrow radiate furrows and wrinkles, with the center umbilicate; white margin about 1.5 to 2 mm. during the growing period; in color blue green shades such as 367, 372, then dark yellow-green 294, 269, 274, to very dark orange brown shades such as 115, 119, in old cultures; odor strong, suggestive of Actinomyces; reverse in orange yellow shades such as 161, 157, 108-110,

a red orange about 84; conidiophores 50 to 200, or 300 $\mu$  by 2.3 to 2.8 $\mu$ , frequently with short branches 10 to 20 with secondary penicilli, with walls smooth; penicilli 8 to 25 $\mu$  long, with walls smooth; metulae usually present, unequal and asymmetrically arranged, 8, 19–14 or even 16 $\mu$  by 1.5 to 2 $\mu$  at base enlarging to 2.5 to 4 $\mu$  at the apices, in divaricate verticils of 2 to 5; sterigmata about 7 to 9 by 2 to 2.5 $\mu$ , in verticils of 3, 6 to 12, or even 15, variously shaped, beaked; conidia about 2.2 to 2.8 $\mu$ , delicately but densely denticulate, globose, showing long slender connectives.

Habitat: Species isolated from earth under pines in a forest near Poznan in Poland.

Zaleski places this species in "Aspergilloides Wehmer-Dierckx. Series 1: *A. Corylophilum*." Our notes follow: Type strain growing about equally well at 30° and 20°C. Colonies upon Czapek's solution agar velvety in appearance composed of a close meshed felt, with margin plane white to gray and 1 to 1.5 mm., wide, within more or less radiately wrinkled with center convoluted, sometimes buckled in quadrants, and more or less zonate, usually pronouncedly so at margin in old cultures, with conidial area in gray shades rather than definite green or blue green such as Hathi gray, Storm gray (R. L. II) or deep olive gray; reverse from bright to dark yellow and orange, or even orange brown shades; drops, colorless; odor, faint—more or less characteristic; conidiophores mostly very short; penicillus consisting of main axis and divergent branchlets 10 to 15 $\mu$  long, each bearing a verticil of sterigmata 7 to 8 $\mu$  long with chains of conidia more or less loosely arranged in twisting or coiling columns up to 200 $\mu$  long; conidia varying somewhat but about 3 $\mu$  in diameter, roughened or with dark granules arranged more or less in bands.

248. *P. Kapuscinskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 484, 485; Taf. 55; Zaleski no. 788b.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 24 to 28 mm. in diameter in twelve days, liquefying the gelatine fairly rapidly, velvety, zonate in the marginal areas only, wrinkled cerebriform in center becoming radiate wrinkled toward the marginal area; white margin 1 to 1.5 mm. during the growing period; in color conidial area blue green shades at first 397, 378B, then 366, 367, 373, 374, later green 322, 323, and in age dark shades of yellow such as 173, 143, 148; reverse in shades of orange such as 146, 141, 196, 156, 191, 136; odor none; conidiophores 100, 200 to 400 or up to 700 $\mu$

by  $2.3$  to  $2.8\mu$ , with apex inflated, straight or flexuous simple or scantily branched; penicilli 8 to 10 or 25 to  $28\mu$  long; metulae commonly unequal in length and asymmetrically arranged in the verticil 10, 12 to 18, or even  $22\mu$  long by  $2.5$  to  $3\mu$ , enlarging from base toward the apex which is usually vesicle-like; sterigmata about 8 to 9 by 2 to  $2.5\mu$  in verticils of 6, 8 to 12 or even to 16, with short tubes; conidia  $2.2$  to  $2.5\mu$  even to  $3\mu$ , smooth, mostly globose, occasionally subglobose, developed in chains showing a long slender connective.

Habitat: Species isolated from sandy soil on the seashore of the Baltic in Poland.

Zaleski reports Biourge as placing this form close to *P. hirsutum* Dierckx in the Hemi concentrica of the Section Bulliardium of Biourge which Zaleski rejects and places it instead in "Biverticillium Dierckx Subsec. 4. Cerebriformiter-radiate-undulata." Our notes follow: Type strain growing fairly well about  $20^{\circ}\text{C}.$ , very slightly at  $30^{\circ}\text{C}.$ , or above; colonies on Czapek's solution agar about  $20^{\circ}\text{C}.$ , restrictedly growing 15 to 20 mm. in diameter in twelve to fourteen days appearing velvety but with very thin felt at base, buckled and radiately wrinkled sometimes in quadrants, with conidial area gray green, deep olive gray (Ridgway LI.) at first, becoming drab in age (brownish drab Ridgway XLV.) with mycelial hyphae, and marginal 3 mm. white, showing stolon-like hyphae reaching beyond the submerged mycelium and reentering the substratum; hyphae very delicate; reverse in pale orange shades; drops, small, colorless, well-distributed over the conidial area; conidiophores; penicillus partly monoverticillate, mostly variously showing one-branch, a group of branches at the first septum, or a group or sterigmata around the main axis at the first septum; branches (or metulae?) 12 to  $15\mu$  long; sterigmata 8 to 10 by 2 to  $2.5\mu$  with conidial chains divergent; conidia  $2.5$  to  $3\mu$ , with delicate roughenings or spinulosity seen only under oil immersion.

Culture no. 5010.12 received from Baarn in July, 1928, tallies well enough with Zaleski's description to be accepted as type.

*Conidia elliptical.*

*P. briosii* Carbone. Atti Ist. Bot. dell Università Pavia, Ser. II, 14 pp, 303-308, 321, Tav. XII, figs. 1 and 8.

Translation (C. T.) of Latin diagnosis: "Colonies ochroleucus, with areas of aeruginous-green; sterile hyphae hyaline scarcely septate,  $1.5\mu$  diam.; fertile hyphae (conidiophores) very rarely or not septate, bearing

1 branch or none, 13.5 to 24 by  $3\mu$ , bearing one verticil of sterigmata 11.2 by  $1.5\mu$ , at the apex and sometimes also around the base of the branch or around the hypha; conidial fruit green,  $31\mu$  in diameter by  $54\mu$  long; conidia in chains, olivaceous, smooth, ovate-acute, or lemonshaped,  $3.5$  to  $4\mu$  by  $2.7$  to  $3.2$ ."

Habitat: Upon sausages.

Carbone's figures are obviously schematic, but figure 8 showing a verticil of four sterigmata shows them clustered at the base and sharply divergent at the apex also the conidia figured are apiculate at one or both ends as in newly formed spores of this series. This conclusion is in harmony with fig. 1 in which the conidial chains are represented as divergent.

## CHAPTER XVIII

### THE ASYMMETRICA-FUNICULOSA

*Section 5. Asymmetrica-funiculosa.* Species in which trailing aerial ropes (funicles) of hyphae are evident under the microscope at least at the margin of the growing colony and usually throughout the colony.

This section includes species whose affinities with the Section *Lanata-Divaricata* are so evident that separation seems hardly justifiable. In some cases the basis of separation is scarcely evident. In general, however, the tendency of hyphae to become aggregated into prostrate, trailing and anastomosing ropes or bundles is readily determinable under the lower magnifications of the compound microscope and is often visible under a good hand-lens (such as 10X). The basis of separation thus appears to be workable for most species. The species brought together then fall into two general subsections (1) *Funiculosa-divaricata* in which the penicillus is divaricate as seen in the preceding section and (2) *Funiculosa-typica* in which the more compact brush or broom type of penicillus is at once evident.

#### *Key to species included*

- I. Penicilli with metulae or branches divaricate.....II.
- I. Penicilli with the compact type of penicillus.....XV.
- II. Conidial chains parallel or diverging not in columns.....III.
- II. Conidial chains forming columns.....X.
- III. Conidial rough or echinulate.....IV.
- III. Conidia smooth 2 to  $2.5\mu$  with overgrowth of funiculose hyphae in age; not rosy in growing areas.....*P. Rivolii* Zal., 235.
- IVa. Colonies floccose-funiculose, gray-green, reverse in purple drab shades; conidia 4 by  $3\mu$  rough....*P. Daleae* Zal., 260.
- IVb. Colonies bluish gray green; reverse white or bluish, conidia rough 3 to  $4\mu$ .....*P. acidoferum* Sopp, 261.
- IVc. Colonies in close textured felts about  $300\mu$  thick, olive-buff or with a trace of green; reverse colorless to rosy; conidia 2.5 to 3.5, rough.....*P. Krzemieniewskii* Zal., 262.

- IVd. Colonies loosely floccose-funiculose white, gray or greenish, more or less zonate; conidia mostly 3 to 4 by 2.5 to 3 $\mu$  almost smooth.....*P. piscarium* Westling, 424.
- V. Conidia smooth in our cultures.....VI.
- VIa. Colonies close textured felts composed of fine hyphae bluish green to pale green to brown; reverse pale yellowish to greenish or uncolored; conidia 3 to 4 by 2 to 3 $\mu$  figured as delicately echinulate but smooth in our culture of Biourge's type.....*P. ochrochloron* Biourge, 263.
- VIb. Colonies bluish green to green shades; becoming slowly reddish; reverse in yellow to ochraceous or violet shades; conidia 2.4 to 3 $\mu$ .....*P. janthinellum* Biourge, 234.
- VIc. Colonies blue gray to brown in age, often showing margin green and center brown; conidia about 2.5 $\mu$ .....*P. glauco-ferugineum* Sopp, 264.
- VIId. Colonies forming broadly submerged felts slowly developing a surface network of trailing hyphae and ropes of hyphae; conidial areas pale green passing to gray or drab; conidia 2 to 3 $\mu$ .....*P. Godlewskii* Zal., 265.
- VIe. Colonies lanose-funiculose, white with pale greenish conidial areas, becoming pale shades of brownish; conidia 2.2 to 3 $\mu$ .....*P. Thomi* Zal., 266. Synonym probably of *P. intricatum* Thom.
- VIIf. Colonies lanose-funiculose white, greenish gray, gray; conidia 2.5 to 3 $\mu$ .....*P. intricatum* Thom, 267.
- X. Conidial chains forming compact diverging columns. Colonies showing a network of hyphae and ropes of hyphae; blue green; reverse yellow; conidiophore walls rough.....XI.
- XI. Colonies blue green; conidia globose 3 to 3.5 $\mu$ .....*P. Howardii* Thom, 268.
- XI. Colonies white to flesh color or near avellaneous; conidia 4 by 2 $\mu$  to 5 by 3 $\mu$ .....*P. Putterilli* Thom, 269.
- XV. Species with metulae or branches of the penicillus aggregated to form the brush or broom type of penicillus (an arbitrary group).....XVI.

- XVI. Colonies a brilliant parrot green in the growing stage.....*P. psittacinum* Thom, 270.
- XVI. Colonies in other shades.....XVII.
- XVII. Colonies zonate.....XVIII.
- XVIII. Colonies deeply floccose in center thinning toward margin; fulvous below.....*P. griseo-fulvum* Diereckx, 273.
- XVIII. Colonies floccose, zonate, reverse ranging from colorless to varying zones of yellow to orange....*P. terrestre* series, 274.
- XVIII. Colonies floccose bluish green with bundles more funiculose than fasciculate in the marginal area.....*P. solitum* Westling, 275.

\* *Conidial chains parallel or divergent not forming compact columns.*

260, 261, 262. The *P. Daleae-acidoferum* series.

*P. Daleae*, *P. acidoferum*, and *P. Krzemieniowskii* present a series of rough-spored soil forms comparable to the *P. nigricans-Janczewskii* series in the *Lanata-Divaricata* (244-248). These are definitely funiculose species. Perhaps further study will put them all together in a single series as we had them once but in our cultures the ropes of hyphae proved a means of separating the series which otherwise seemed to run together.

*P. Daleae* Zaleski. In *Bul. Acad. Polonaise Sci.: Math. et Nat.* Ser. B, 1927, pp. 495, 496; Taf. 57; Zaleski no. 1317a.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 24 to 26 mm. in diameter in twelve days, liquefying the gelatine tardily but completely, velvety or somewhat closely subfloccose, zonate only indistinctly and in the outer area, with the whole central area thrown into broad regularly radiate wrinkles, with the very center somewhat depressed and showing a few uncolored drops; white marginal zone 2 to 3 mm. wide; in color conidial areas at first blue green shades such as 371, 372, becoming dark yellow green shades such as 273, and later dark orange brown such as 168, 164, 138, 139; reverse at first in orange yellows such as 171, 166, 157 to 133, 138, later becoming red orange 84, 88, 92, 97; odor none or weak; conidiophores 10 to 200 or 300 by 2 to 2.5 $\mu$ , with apex more or less enlarged or inflated, com-

monly unbranched, occasionally with short branches, flexuous, varying greatly in length, erect or ascending; penicilli mostly 10 to 12 $\mu$ , less frequently 25 to 30 $\mu$  or 40 $\mu$  long, with walls smooth; metulae 8, 10 to 20 or 24 by 2.5 to 3 $\mu$ , in groups of 2 or 3, commonly unequal and irregularly arranged, with apices commonly inflated; sterigmata about 9 to 10 by 2.5 to 3 $\mu$ , commonly in verticils of 3, 5 to 10 or 12, sometimes occurring singly; conidia 2.5 to 4 by 2.5 to 3 $\mu$ , varying considerably in size, coarsely denticulate, ovate elongated or subglobose.

Habitat: Species isolated from soil under pine near Poznan, Poland.

Zaleski notes that Biourge in examining this species placed it near to or identical with *P. janthinellum* Biourge. The species grows poorly on neutral Raulin gelatine. He puts it among the "Aspergilloides Wehmer-Dierckx Series 1: A Corylophilum." Our notes follow: The type strain growing well at both 20° and 30°C. Colonies upon Czapek's solution agar at 20°C., spreading fairly widely (30 mm. in diameter in seven days), floccose with some funiculose or fasciculate hyphae (more deeply floccose in slanted tubes than in petri dish cultures), gray green with white marginal areas 2 to 3 mm. in width during the growing period (at 30°C. pitted mycelial mass thinner, radiately wrinkled and wanting in green color); reverse in areas purple drab (Ridgway XLV.); drops in the central area, colorless; conidia coarsely roughened with winding colored bars elliptical to subglobose 4 by 3 $\mu$ .

261. *P. acidoferum* Sopp. Monogr., pp. 188-189; Taf. XXI, fig. 146; Taf. XXIII, fig. 34. 1912.

Colonies on meat-peptone-sugar-gelatine, gray blue, in age dark olive green, fibrous, with wrinkled surface growth; hyphae delicate; reverse white or almost bluish; gelatine liquefied; odor of onions; conidiophores arising from creeping or trailing hyphae, short or fairly long, branching with each branch becoming a diverging monoverticillate secondary conidiophore or bearing a terminal verticil or umbel 8 to 10 $\mu$  diverging metulae each bearing a verticil of sterigmata and spore chains; sterigmata not described; conidia echinulate, globose, 3 to 4 $\mu$  in long axis; perithecia not found.

Species found in earth in West Norway. Cultures grew best at 20°, with minimum at 3° and maximum at 35°C., and grew well in all the common media tested. Conidia remained viable more than three years.

Gilman and Abbott (on p. 295, fig. 30) identified a culture from Utah soil as Sopp's *P. acidoferum* (our no. 4914). In our cultures this species produced colonies bluish gray, to gray, floccose to ropy, 500 to 1000 $\mu$



deep, more or less buckled and deeper in center; reverse yellow to reddish brown; odor evident; conidia rough 3 to  $4\mu$ . Gilman and Abbott's description reads (compare fig. 57.): Colonies on Czapek's or bean agar slowly spreading, cottony or closely floccose; surface olive gray; reverse orange buff; pigment diffuses into the medium. Aerial hyphae abundant, smooth; hyaline, creeping. Conidiophores arise from aerial hyphae as short side branches up to 40 or  $50\mu$  long, unbranched, or once or twice branched at the apex; each branch bears a terminal head of conidial chains, which are loosely columnar, up to 75 or  $100\mu$  long, and usually with 5 to 10 chains in each head; apex of conidiophores slightly inflated; phialides  $6.5$  to  $8.5\mu$  long by 2 to  $3\mu$  thick. Conidia globose, light green, delicately rugulose under oil,  $2.5$  to  $3.5\mu$  in diameter. Elements of conidial fructification pitted. Sopp (44) gives  $3-4\mu$  for conidia.

From soil: Norway (44).

United States: Utah.

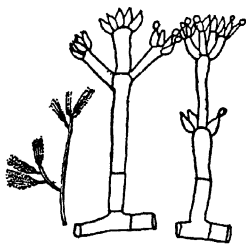


FIG. 57. *P. acidoferum* after Gilman and Abbott (fig. 30): Detail and sketch

This species is closely related to *Penicillium rubens* Biourge (9). *P. acidoferum* has smaller conidia and phialides and the elements of the fructification are rough, whereas those of *P. rubens* are smooth.

262. *P. Krzemieniewskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 490, 491, 492; Taf. 56; Zaleski no. 1362.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 28 to 30 mm. in diameter in twelve days, liquefying the gelatine slowly but completely; velvety, plane and azonate or indistinctly zonate in the outer area; areas within thrown into broad irregular wrinkles; white margin 1 to 1.5 mm. during the growing period; in color conidial areas green shades such as 347, 346, 348, becoming dark orange brown shades such as 143, 147 in age; reverse in light orange yellow shade such as 171, 166, 157, 162, becoming very dark purplish at

times in age; odor none; conidiophores 50 to 300 or 400 $\mu$  by 2.5 to 3 $\mu$ , with apices inflated, sometimes unbranched, mostly branched more or less irregularly; penicilli when simple 10 to 12 $\mu$ , mostly 20 to 30 or 35 $\mu$  long, with walls smooth with anomalies of gigantic sterigmata and thyrsiform masses common; metulae 10, 12 to 20, or 25, by 2.5 to 3.5 $\mu$ , commonly unequal in length and asymmetrically arranged in verticils of 2, 4 or 6; sterigmata about 9 to 11 by 2.5 to 3.5 $\mu$ , in verticils of 5 to 12 or 15, or few or even singly, straight or incurved, mostly with long slender tubes; conidia 2.5 to 3.5 or 4 $\mu$ , delicately but densely denticulate, subglobose, showing a distinct connective.

**Habitat:** Species isolated from sandy soil under pines near Poznan, Poland.

Zaleski finds that the characters given do not place this species satisfactorily; recognizes a possible relation to the *Thyrsiferentes* but places it in "*Aspergilloides* Wehmer-Dierckx Series 1: *A. corylophilum*." Our notes follow. Type strain growing more rapidly at 30°C., or above, than at 20°C.; colonies upon Czapek's solution agar forming close textured felts with surface hyphae partly funiculose, about 300 $\mu$  in thickness, buckled and radiately wrinkled, more or less zonate, in most cultures showing at first large water soaked areas, pale olive buff (Ridgway XL) with only a trace of green in the color; reverse colorless or with a slight rosy or reddish tinge at first, becoming brown in some cultures and more pronouncedly red on wort; conidiophores about 2 to 3 $\mu$  in diameter with walls smooth or showing a trace of granulation mostly on the inner face of the cell wall; penicillus either a single verticil of sterigmata or a terminal group of monoverticillate branches with sessile sterigmata occasionally at lower septa of the main axis; sterigmata 7 to 10 by 3 $\mu$  conidia 2.5 to 3.5 subglobose, rough with winding color bars.

*P. ochro-chloron* Biourge. Monogr. La Cellule 33: fasc. 1, p. 269-270; Col. Pl. X, Cart. 192; Pl. XVII, fig. 100. 1923.

Colonies on wort gelatine, spreading, more or less wrinkled, at first bluish green, then green at length brown; coremia none; reverse pallid or sordid yellowish, then yellow, with brown spots or patches; odor, none; conidiophores 2.8 to 3 $\mu$  in diameter with walls rugulose, arising from creeping hyphae, figured as very short; penicillus 20 to 50 $\mu$  long, all walls squamulose rugulose, figured as a simple verticil of sterigmata, or a one-sided verticil of metulae, or with one or more branches from lower nodes; metulae 11 to 13, or even 20 to 30 by 2 to 3 $\mu$ ; sterigmata 9 to 11 by 3 to 4.5 $\mu$  in groups of 3 to 6; conidia 3 to 4 by 2 to 3 $\mu$ , elliptical to subglo-

bose; figured as delicately echinulate; Biourge's no. 192 was received from him in September, 1927; upon Czapek's solution agar colonies composed of fine hyphae, closely felted—floccose, with tufted uneven surfaces, with abundant trailing hyphae and ropes of hyphae, white becoming pale greenish in areas of abundant conidium production; reverse pale yellowish or greenish yellow or scarcely colored; odor none; drops not seen; conidiophores from very short branches of aerial hyphae to  $200\mu$  by about  $2\mu$ , in our preparations smooth walled; penicillus monoverticillate or with one or more unequal branches, each having a verticil of few, closely packed sterigmata 7 to  $10\mu$  long, bearing conidial chains diverging, parallel, or sometimes almost in a column up to  $100\mu$  long; conidia uneven in size 3 to 4 by 2 to  $3\mu$  and so far as seen smooth or nearly so.

264. *P. glauco-ferugineum* Sopp. Monogr., pp. 152–153, Taf. XVII, fig. 116, Taf. XXII, fig. 9. 1912.

Colonies on meat peptone sugar gelatine, blue-gray, in age brown, often showing margin green and center brown, with heavy mycelium wrinkled or buckled, with aerial hyphae in bundles and ropes bearing the conidiophores as branches; hyphae comparatively fine; reverse gray brown, to brown, or greenish brown; odor not definite except a weak arsenic odor on potato plugs which are colored almost black; conidiophores short, slender, often unbranched monoverticillate, again (from figures) producing verticils of metulae or even an additional branch at a lower level; metulae when present with enlarged apices; sterigmata very short; conidia globose, smooth  $2.5\mu$ ; perithecia (? sclerotia) greenish, found once but not described, hence doubtful.

Species found in earth, colonies showed an optimum temperature of  $20^{\circ}$ , with minimum at  $8^{\circ}$  and maximum at  $33^{\circ}\text{C.}$  and grew well upon the usual media.

Culture no. 4906 IV isolated by C. E. Burnside from a dead honey bee, reproduces Sopp's description so well as to justify acceptance under the name. Upon Czapek's solution agar, colonies blue-grey to rusty brown in age, with thin floccose slightly ropy surface, with conidiophores partly borne upon aerial hyphae, partly from submerged hyphae, mostly 100 to  $200\mu$  long, about 1.5 to  $2.5\mu$  in diameter, with walls pitted, showing irregularly disposed swollen cells, sometimes in the conidiophores, again the branches or sterigmata, and often whole series of swollen cells in the submerged mycelium; penicillus simple and monoverticillate or a one-sided verticil of branches or metulae.

*P. janthinellum* Biourge (no. 234), by description would fall at this point. Without seeing the type we are convinced that the name belonged to some one of the series of strains constantly encountered in soil and showing every gradation from simple floccosity to floccosity mixed with ropiness and many of them characterized by sooner or later producing hyacinth shades in reverse especially in soil extract agar, but usually also in Czapek. All of them were therefore put among the *Lanata-divaricata* in Chapter XVII.

265, 266, 267. The *P. Godlewskii-intricatum* series.

White to faintly greenish gray strains, with trailing ropes of hyphae and scanty development of monoverticillate-divaricate and biverticillate divaricate penicilli, are not uncommon in soil plates. *P. intricatum* was isolated in Connecticut; a nearly related form was sent in by Miss McLennon from Australia; Zaleski obtained his organisms in Poland. Separation among them upon the data we have is difficult or doubtful.

265. *P. Godlewskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 466, 467; Taf. 45 and 49; Zaleski no. 1432b.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes; slow growing becoming 24 to 26 mm. in diameter in twelve days with symmetrically radiate wrinkles 2 to 3 mm. wide and 1 to 2 mm. in height, and the center raised as a cushion or umbo often above the wrinkles, with gelatine tardily but completely liquefied; conidial area velvety or slightly floccose, azonate; margin plane, sordid-white, 2 to 3 mm. wide, in color at first white for several days then with central conidial areas blue green (C.d.C. 397, 372, 396, 371) becoming orange brown in age 147, 148; reverse pale yellow 0146, and 146, 153B, 128B; drops few, central; odor none; conidiophores as short as  $100\mu$  mostly 200 to 400, (occasionally 600) by 2 to  $3\mu$ , straight or flexuous, with apices vesicle like; penicillus from 10 to  $25\mu$  long, with much variation in structure; branches ordinarily absent; an occasional distant branch is indicated by the figure; metulae about 10 to 14 by  $2.5$  to  $3\mu$ , with apices flattened or vesicle-like, straight or incurved, absent or unequal, symmetrically or asymmetrically arranged in verticils of 2 to 8; sterigmata about 7 to 8 by  $2.2$  to  $2.5\mu$ , commonly 6 to 12 in the verticil, with short acute tubes; conidia 2 to  $2.5\mu$  (even 2.8), smooth, globose, showing a connective, and long adhering in the chains.

Habitat: Species isolated from earth under pine trees in square of

652 of the forest "Puszcza Bialowieska" in Poland. Zaleski puts it in "Biverticillium Dierckx subsec 3. Radiate-undulata."

Study of the description and figures does not warrant assignment to *Biverticillium* in the sense of this book. Our notes follow: Type strain growing very poorly at 30°C., fairly well at 20°C.; colonies upon Czapek's solution agar about 20°C., forming felts mostly submerged often sodden or wet looking, showing cerebriform or convoluted wrinkling in the central area, and radiate wrinkling toward the margin, with surface growth often scanty or bristly at first, when well developed consisting of a network of trailing hyphae and ropes of hyphae (funiculose), with marginal 2 mm. white, conidium bearing band or zone of pale green, passing in the older areas to gray, pale drab and ecru drab (Ridgway); reverse uncolored or indefinitely colored; conidiophores commonly branching from aerial hyphae, 50 to 500 $\mu$  long; penicilli either a single verticil of sterigmata or a group of 2 to several divergent unequal branches mostly fairly short, with sterigmata and conidial chains divergent; sterigmata 6 to 7 $\mu$  long; conidia colorless 2 to 2.5 $\mu$  in diameter, showing connectives.

Culture no. 5010.31 received as type from Baarn in July, 1928, complies nearly enough with Zaleski's description to establish a presumption of identity. In our cultures, the species falls in a series with *P. intricatum* Thom.

266. *P. Thomi* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 492, 493; Taf. 56; Zaleski no. 1589a. Not *P. Thomii* Maire.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, rather quickly growing becoming 33 to 35 mm. in diameter in twelve days, liquefying the gelatine slowly but completely, lanose-composed of dense hyphae up to 5 mm. long, merging at the margin into a fimbriate white area 3 to 4 mm. wide, and including anastomosing ropes of hyphae which bear conidiophores as branches; conidial area pale greenish at first then such shades as 372, 322, with the green finally fading out leaving shades of orange brown such as 137, 121, 103C; reverse in orange yellow and orange shades such as 171, 162, 161, 129, 109; odor none; conidiophores from very short to 100 or 200 by 2 to 2.5 $\mu$ , usually flexuous, borne as branches of long (2 to 4 mm.) erect or trailing aerial hyphae and ropes of hyphae; penicilli 8 to 12 $\mu$  when simple, 15 to 28 $\mu$  when branched, with walls smooth; metulae 8, 10 to 16, or 20 by 2.5 to 3 $\mu$ , divaricate varying in length and arrangement in groups of 2 to 5, usually enlarging upward, or inflated at the apex; sterigmata about 7 to 9 by 2 to

2.5, very variable in size and appearance, short and inflated or straight, commonly with long tubes or beaks, verticils of 5, 8 to 12 or 15; conidia 2.2 to 2.8 or  $3\mu$ , smooth, more or less globose, showing connective in the chain but with the chains easily breaking down.

Habitat: Species isolated from soil under pines in the forest "Puszcza Bialowieska" in Poland.

Zaleski placed this species with the series "Corylophilum" as the section with characters nearest to this form but believed it should be made the type species of a new series among the *Aspergilloides* Wehmer-Dierckx Series 1: *A. corylophilum*." Our notes follow: Colonies upon Czapek's solution agar, spreading broadly, rather deeply floccose and partly funiculose, consisting of delicate hyphae 1 to  $2\mu$  in diameter, forming a fine cottony mass white to slightly cream or grayish, without definite green color, since conidium production is very scanty; reverse colorless to cream or at times to pale yellow shades; conidiophores found as very short branches of aerial hyphae; penicilli consisting of single verticils of few sterigmata with divergent chains of conidia; sterigmata about 10 by  $2.5\mu$ ; conidia 2.5 to  $3\mu$  in diameter;

Culture no. 5010.26 received as type from Baarn in July, 1928, lacks the green colors of Zaleski's description. Its identity as type is therefore doubtful. It resembles *P. intricatum* Thom closely enough to be placed under that name. Zaleski's specific name, *P. Thomi*, is antedated by Maire's *P. Thomii* which is a very different species hence culture no. 5010.26 may be assigned as *P. intricatum* leaving its identity with Zaleski's species questionable.

The various strains assigned here differ somewhat in cultural reactions but comply with a general morphological picture closely enough to be left together until some one by comparative study can establish their real relationships.

267. *P. intricatum* Thom, U. S. Dept. Agr. Bur. Anim. Bull. 118: 75-76, fig. 31. 1910.

Colonies upon gelatin or bean agar, zonate in some cultures white, gray greenish gray, when old gray or almost smoky becoming a mass of interwoven hyphae and ropes of hyphae 1 to 3 mm. in thickness; reverse of colony and substratum not colored in bean agar, more or less sulphur yellow or even brownish in sugar media; conidiophores sometimes terminal, more commonly branches of aerial hyphae 30 to  $50\mu$  in length; penicilli 50 to 100 up to  $140\mu$  in length, or much longer in old sugar cultures, consisting of simple verticils of sterigmata, or of 1 to 3 verticils upon

divergent branchlets, or of branchlets and sterigmata in the same verticil; sterigmata 8 to 10 by 2 to  $2.5\mu$ , few (4 to 10) in each verticil, bearing more or less divergent chains of conidia frequently aggregated into a loose column; conidia elliptical to globose, hyaline or pale greenish,  $2.5$  to  $3\mu$  in diameter, smooth, thin walled, granular within, remaining in chains in fluid mounts; colonies alkaline to litmus, not liquefying gelatin.

Found in cultures from soil, Storrs, Conn., by Prof. W. M. Esten, 1907.

Color white or greenish gray—not green, grayish to brown or drab; reverse and medium uncolored or sulphur-yellow in some media; litmus reaction strongly alkaline. Potato and bean agar, typical. Potato plugs, weak growth, not adapted to this species. Cohn's solution, weak growth, yellowish-green colonies.

*\*\*Conidial chains forming compact columns.*

*Colonies with aerial hyphae in ropes; each metula producing a long, diverging monoverticillate column of conidial chains.*

- a. Colonies blue green; conidia globose 3 to  $3.5\mu$ .....*P. Howardii* Thom, 268.
- b. Colonies white to flesh color; conidia 4 by  $2\mu$  to 5 by  $3\mu$ ...*P. Putterillii* Thom, 269.

The two species described here seem necessary to account for the record of described forms. It is regrettable to find such description necessary after the cultures have been lost.

**P. Howardii** Thom, n. sp. Type no. 4659, no longer viable.

Colonies upon Czapek's solution agar, floccose-funiculose blue green; reverse in pale yellow shades (luteus); conidiophores rough; penicilli consisting of 3 to 4 metulae 10 to  $13\mu$  long with very long dense divergent columns of conidial chains; sterigmata 6 to  $7\mu$  long, closely packed, with apices acute; conidia globose 3 to  $3.5\mu$ , dark green;

"Found upon an oak block which had been repeatedly wet with mercuric chloride solution."

Habitat: Providence, Rhode Island; contributed by Dr. N. O. Howard in culture, 1923.

**P. Putterillii** Thom, n. sp. Type no. 4658.3.4 not now viable.

Colonies upon Czapek's solution agar floccose funiculose with ropes of

hyphae forming a thin tufted close lying layer, with hyphae coarse, in color white to flesh color, cartridge buff, almost avellaneous (Ridgway); reverse colorless or yellowish cream; conidiophores usually less than 100 by 3 to 5 $\mu$  with walls rough borne as short 1 to 2-celled branches from ropes of hyphae; penicilli variously a main branch with or without a verticil of 2 to 4 branches, then 1 or several verticils of metulae 8 to 12 by 3 to 4 $\mu$  with walls rough; sterigmata 10 to 12 by 2 $\mu$ , closely parallel in the verticil; conidia cylindrical to elliptical 4 by 2 $\mu$ , to 5 by 3 $\mu$  colorless as seen under high magnification, in chains massed into long columns.

Habitat: Isolated by F. M. Putterill in his studies of the shipping and storage of fruit in the boats and warehouses at Cape Town, South Africa, 1922.

*Funiculose species with the penicillus of the Fasciculata or the Lanatypica.*

270. *P. psittacinum* Thom, n. sp. Type culture no. 4733.12 from Biourge.

Synonym: *P. aureum* Corda in Biourge Monogr. La Cellule 33: 111-114, Col. Pl. I; Cart. 144; Pl. I, fig. 2. 1923.

See Biourge's description of the same organism as *P. aureum* no. 271.

Colonies upon Czapek's solution agar slowly spreading about 5 to 6 cm. in diameter in ten days and forming a tough felt up to 1 mm. deep upon the surface of the agar, broadly but not very distinctly zonate, and often radiately wrinkled in the outer areas, with broad white margin, conidial areas in the growing period showing a striking shade of parrot green near Malachite green (Ridgway XXXII) or psittacinus, becoming shades of olive gray and overgrown with a thin web of aerial hyphae in age, in which anastomosing ropes of hyphae are abundant; marginal areas during the growing period showing partly funiculose partly fasciculate hyphae; in radial section, fasciculation often appears to be general in the deeper areas; reverse colorless or with pale shades of yellowish orange; odor indefinite; conidiophores with walls pitted, about 4 $\mu$  in diameter, either very short branches of trailing or ascending hyphae; or terminal segments of such hyphae; penicilli asymmetrical consisting of main axis with or without one or more branches commonly one septate and monoverticillate, appressed when present; metulae 10 to 12 $\mu$ ; sterigmata 10 to 12 by 2 $\mu$ , closely packed, few in the verticil; conidia globose,



colorless,  $4\mu$ — or a little larger, marked by points in thick spots in wall— spinulose as described by Biourge.

271. *P. aureum* Corda. In Biourge Monogr. La Cellule 33: 111–114, Col. Pl. I; Cart. 144; Pl. I, fig. 2. 1923.

Synonym for *P. psittacinum* Thom, q.v.

Colonies on wort gelatine zonate, with margin thin white or pale orange (?), mycelium golden yellow, with conidial areas at first parrot green then brownish olive, coremia rarely produced; odor moldy; taste objectionable; conidiophore  $3.5\mu$  in diameter, smooth, penicillus long (included in section with penicillus 60 to  $150\mu$  long), with branches 12 to 20 by  $3.5\mu$ , figured as a main stalk and irregularly 1-sided branching system, with sterigmata at one or at several levels in different penicilli; metulae 12 to 14 by  $3.5$ , in twos, threes, or fours; sterigmata fusoid large, 9 to  $11.5$  by  $3.5\mu$  (Corda gives no definite data) in twos, threes or fours; conidia globose  $3.5\mu$  in diameter, deciduous (Corda figures them definitely as elliptical), figured as occasionally sparingly echinulate.

Biourge emphasizes the parrot-green of the young conidial area followed by olive in the ripe spores together with yellow colors in the mycelium as determining characters in his recognition of Corda's species. He believes the identity to be established.

Biourge's culture (our no. 4733.12) corresponds fairly closely with his description but *P. aureum* of Corda would suggest to us such a form as *P. herquei* Bainier and Sartory which does not seem to have been known to Biourge, (no. 397).

Since *P. psittacinum* was described at least two more strains have come into our possession which belong very near that species. These vary in detail from Biourge's culture as described but have too many characters in common to make diagnostic separation very practical with what we know now. It is better to indicate that *P. psittacinum* will be found to be the type of a series than to limit it too closely to a strain.

272. *P. griseo-fulvum-terrestre* series.

There is a puzzling series of floccose-funiculose forms of varying depth, broadly zonate especially in the marginal areas, in pale green to gray green, olive green to brown shades in age with the final shade varying with the species or strain, with the reverse of colony colorless, variously zoned or banded with yellow which may fade or pass over through orange yellow shades toward fulvous finding its end point at

different intensities among the various strains. Some of these strains produce a strong and rather disagreeable odor in age. They are apparently mostly soil organisms or at least reach the laboratory in soil or soil contaminated substances. Cultures have been examined from Sweden (Melin), Scotland (Birkinshaw), Texas (Lewis), and various incidental American sources. The positive identity of the strain may be difficult to establish but the group is determinable. Among the record numbers included are: 5034.8, .9, .11, and .62; 5042.91; 5007.67, .69, and .72.

273. *P. griseo-fulvum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88. 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 164-167; Col. Pl. II, Cart. 34; Pl. II, fig. 11. 1923.

Colonies upon wort gelatine hemizonate! floccose, at first pale bluish, then gray reddish, with coremia large 4 to 12 by  $2.5\mu$  in height more or less loose in structure with stalks white or yellowish and reverse at first yellow then fulvous, conidiophore about  $5\mu$  in diameter, with walls smooth; penicillus most commonly about  $30\mu$ , occasionally 50 to  $60\mu$  long, branches more even in size, often showing two 12 to  $16\mu$  and one 18 to  $25\mu$  long in the same verticil, some producing metulae, others in the same verticil producing sterigmata; metulae commonly 6 to 10 occasionally to  $17\mu$  by 2.5 to  $5\mu$ , 3, 4, 5 in the verticil; with apex clavate or vesicle-like; sterigmata 7 to 8 or even to  $11\mu$  by 3.5 to  $4\mu$ , in groups of 2 to 8; conidia globose 2.5 to 3.5 or even  $5\mu$ .

Biourge's no. 34 (our no. 4733.69) on Czapek's solution agar floccose becoming 2-3 mm. deep at center as loosely massed aggregations or tufts almost coremiform, gray green and thinning toward the margin where indistinct zones appear, with the outer 1 mm. submerged; reverse and agar yellowish to reddish orange; no conidiophores and conidial fruit visible in the marginal zones with the microscope.

Biourge suggests a comparison with *Coremium vulgare* Corda which appears to us very doubtful. In his "list onomastique" he cites *P. griseo-fulvum* (Dierckx) Galeotti. No such change of name is possible by rules.

274. *P. terrestre* Jensen. Cornell University Exp. Sta. Bul. 315, p. 486-487, fig. 122. 1912.

Colonies on soil extract agar round, yellowish green; vegetative hyphae 2 to  $6\mu$  in diameter; showing superficial ropes of hyphae; conidiophores 70 to  $375\mu$  by 2 to  $4\mu$ , hyaline, septate, either with 1 or 2 branches near the apex or with branching limited to a terminal verticil of metulae;

branches when present bearing terminal verticils of metulae; metulae 10 to  $15\mu$  long; sterigmata 7 to  $11\mu$  long; conidia described as globose 2 to  $3\mu$ , ( $4\mu$  as seen by us in the type specimen) hyaline, in long chains.

Species found in soil plots at Cornell University 1910–1911; the type in (Cornell) plant pathology herbarium no. 5916 was seen by courtesy of Prof. H. M. Fitzpatrick. The funiculose character of the mycelium was clearly seen together with the general type of branching and chains of conidia about  $4\mu$  in diameter.

A composite diagnosis based upon our observations upon some of the strains is desirable to aid in the recognition of the series: Colonies upon Czapek's solution agar fairly widely spreading, zonate and often deeply ridged especially in older colonies, deeply floccose-funiculose with ropiness readily observed and the mass up to  $500\mu$  deep in no. 5042.135, to 800 to  $1000\mu$  deep in no. 5034.9, or to 1 to 2 mm. deep in no. 5034.8 and 5042.91, or thinning to a fraction of the depth and retaining only traces of fasciculation or ropiness in 5034.18, with white or sterile margin broad during the growing period passing into conidial zones in shades of dull green, slightly bluish green, Gnaphalium green of Ridgway, then toward olive gray shades and brown shades in center and throughout in cultures; reverse showing the zonate effect, with yellow to orange color usually showing along the outer zone lines (transient?) especially under the greener zones of conidial fruiting and in the deeper areas of the agar, and central area paler or even uncolored or only slightly yellowish; odor, strong, like certain mushrooms; conidiophores about  $3\mu$  in diameter, varying greatly in length, with walls faintly pitted as seen under oil immersion objective; penicilli few branches with chains in a loose column at first then tangled in age; sterigmata about 7– $10\mu$  long, occasionally few in number and up to  $20\mu$  long; conidia 3 to  $4\mu$ , mostly about  $3\mu$  to  $3.5\mu$  in long axis, when ripe, when young elliptical about  $3.3\mu$  in long axis, later 4 by  $3\mu$ , finally  $3.5$  to  $4\mu$  smooth.

Strains reported in 5034 series came from Nobel's Explosions Company, Ayrshire, Scotland, in no. 5042 from sugar beet studies Logan, Utah, 1929.

275. *P. solitum* Westling. Arkiv för Botanik 11, pp. 52, 65–67, figs. 3 and 47. 1911. Comparison to *P. puberulum* Bainier suggested by Westling.

Colonies in prune gelatine, somewhat floccose in central areas, bluish green (C.d.C. 363, 367) then green (C.d.C. 333, 338) to blackish green (C.d.C. 349) in age; margin narrow, somewhat floccose, white; reverse

sordid white, or pale yellow; gelatine slowly and partly liquefied; odor weak; conidiophores with walls smooth or nearly so, rarely rough, arising from submerged or creeping hyphae, sometimes very short but usually quite long, 300 to 800 $\mu$  by 4 to 6.4 $\mu$ , with penicillus 60 to 150 $\mu$  long; metulae slightly clavate 11 to 18 by 3.6 to 4.8 $\mu$ ; sterigmata 8 to 9.6 by 3 to 3.4 $\mu$ ; conidia elliptical to globose or less often oval, smooth, commonly 3.6 to 4.6 $\mu$ .

Species found first in a hazel nut in association with *P. tabascens*, later on fruits of *Vanilla planifolia*, and afterward found upon many substrata. Calcium oxalate crystals mostly as needles were enmeshed in the mycelium. Colonies grew well upon all media tested. Westling's culture (our no. 2546) received as this form has been kept for many years and similar organisms have been identified from various substrata. Our notes follow: Colonies in Czapek's solution agar with cane sugar, blue green from no. 396 at margin through 397, to 393 or sometimes 367 passing later to green shades nos. 347, 343 in Klineksieck's Code; floccose, uneven or tufted in central area, spreading broadly with more or less definite ropiness in the broad white border; reverse tinged with yellow; odor fragrant; conidiophores smooth; conidial fructification once or twice verticillately branched, with branches appressed; sterigmata 8.5 to 10 $\mu$  by 2 to 3.5 $\mu$ ; conidia smooth, globose or broadly elliptical, 3.3 to 3.6 even 4 $\mu$  in diameter; granular within, persistent in chains when mounted.

Gelatin in water liquefied beneath the colony, dull green, without odor, with colony blue green no. 367.

Bean agar produced colonies pale blue green at first C.d.C. no. 0371, passing quickly to dark shade no. 375, zonate, thin growth, producing conidia 3.5 to 4.5 by 3 to 3.5 $\mu$ , smooth, homogeneous, showing prominent connective.

Biourge, Monogr. La Cellule 33: pp. 114-116, Col. Pl. I, Cart. 3, Pl. I, fig. 3, 1923, regarded this species as certainly *P. glaucum* as understood by Dierckx (1901). Colonies on wort gelatine zonate, floccose, blue green (C.d.C. 353, 363, 378), without coremia, no overgrowth; reverse orange yellow but no diffused pigment; drops uncolored; liquefaction slow, incomplete; odor moldy, ethereal, strong; conidiophore about 4 $\mu$  in diameter; penicillus freed from conidia 40 to 100 $\mu$  long, figured as main stem and branch irregularly producing metulae and sterigmata at various levels; branches 20 to 24 by 4 $\mu$ ; metulae 12 to 22 by 3.5 to 4 $\mu$ ; sterigmata 9 to 10.5 by 3.5 to 4 $\mu$  in three's or four's; conidia globose or subglobose, 4 to 4.8 by 3.8 to 4.5 $\mu$ , persisting in chains.

Biourge's culture no. 3 (our no. 4733.114) appears to be nearly if not identical with no. 2546.

## CHAPTER XIX

### THE FASCICULATA

*Asymmetrica* Section 6. *Fasciculata*. Species are characterized by the aggregation of part or all of the conidiophores into erect bundles or fascicles. Appearances range from colonies showing rudimentary bundles of conidiophores giving a granular, tufted or rough appearance to the white margin, or others with definite coremia mixed with simple conidiophores, to a few species with all or nearly all their conidiophores aggregated into more or less sharply marked coremia.

*Ascospores*. Perithecium formation has been described by Schwartz for one member of this group, *P. italicum* Wehmer (see no. 326). Brefeld's description of sclerotium and perithecium formation in *P. glaucum* may or may not belong here. On account of lack of any basis for identification we have put Brefeld's *P. glaucum* among the undeterminable species in Chapter XXV. Section includes nos. 285 to 354.

Gradually we have come to believe that descriptions in which the colony is described as "granular," "mealy," "tufted," as well as the variations upon such words as coremia, coremiform, flabelliform, Isariaeform are all more or less descriptive of the appearance given to the surface of the growing colony by the fasciculation of its conidiophores. Necessarily as in every attempt made at separating such species as these, questionable forms are encountered. In some of these the superficial mealiness or roughness of the surface is evident under the handlens but definite bundles of conidiophores are scarcely seen when the compound microscope is used. Hence doubts of allocation are certain to arise. In such cases arbitrary assignments have been made and have been based upon what seemed to be the more useful diagnostic characters. Cross references have therefore been introduced into the Key of the chapter to reduce confusion as much as possible.

The Key to species is introduced first, followed by the analysis of the sub-sections as an attempt to show real relationships.

#### *Key to the species assigned to the Fasciculata*

- I. Species producing sclerotia or perithecia. .... II.
- I. Species not reported as producing sclerotia or perithecia. .... III.

- II. Sclerotia predominating at temperatures of 20 to 30°C.; fasciculate at temperatures of 15° or lower.....*P. gladioli* Machacek, 285.
- II. Sclerotia or perithecia occasional or under special conditions; fasciculation regularly present.....*P. italicum* Wehmer, 326.
- III. Colonies with simple conidiophores and fascicles closely mixed, but with simple conidiophores predominating.....IV.
- III. Colonies with most or all of the conidiophores in fascicles or in definite coremia.....V.
- IVa. Colonies gray, with young colonies stellate.....*P. Schneggii* Boas, 330.
- IVb. Colonies in blue green shades.....Sub-section *Aeruginosa* VI.
- IVc. Colonies in bright green or yellowish green shades.....Sub section *Viridicata* X.
- IVd. Colonies in dull to dark gray green or glaucous shades.....Sub-section *Glaucata* XI.
- V. Surface growth consisting of crowded fascicles mostly small with simple conidiophores among them.....Sub-section *Coremiella* XXV.
- V. Coremia evident and standing separately with simple conidiophores reduced or absent.....Sub-section *Coremia* XXX.
- VI. Species in blue green shades (on Czapek): Conidia globose or subglobose.....VII.
- VI. Species in blue green shades: Conidia mostly elliptical.....VIII.
- VII. Conidia mostly about 3 $\mu$ , or less frequently larger, colonies azonate or hemizonate in age.....*P. cyclopium* and related strains, 286. Compare *Coremiella* XXV.
- VII. Conidia about 4 $\mu$ , with individuals much larger and often elliptical; colonies markedly zonate in age.....*P. flavoglaucum* Bourse, 288.
- VIIa. Perhaps nearly related.....*P. conditaneum* Westling 289.

- VIII. Colonies zonate and fasciculate at margin; appearing as a deep dense felt in central area.....*P. janthogenum* Biourge, 290.
- VIII. Colonies zonate.....
- VIIIa. Zones broad; reverse yellow to maroon.....*P. martensii* Biourge, 291.
- Zones broad; reverse scarcely yellowish.....
- VIIIb. Zones narrow; colonies 100 to 200 $\mu$  deep; reverse colorless to purplish or vinaceous.....*P. majusculum* Westling, 292.
- VIIIc. Zones narrow crowded; reverse yellow to purplish or nearly black.....*P. Johannioli* Zal., 293.
- VIIIId. Zones narrow crowded; reverse in deep orange shades. Zones 1.5 mm. wide; persistently blue green; reverse slowly maroon.....5042.128.
- X. Colonies bright green or yellowish green.....*P. viridicatum* series X.

*Abstract of possible lines of separation within the P. viridicatum series*

Xa. Conidia subglobose:

1. *P. musae* Weidemann, 296.  
Conidia 2.2 to 2.8 by 2 to 2.3 $\mu$ .
2. *P. olivino-viride* Biourge, 297.  
Conidia 2.5 to 3.5 $\mu$ .
3. *P. viridicatum* Westling, 293.  
Conidia 3 to 3.8 $\mu$ .
4. *P. verrucosum* Dierckx, 299.  
Conidia 2.8 to 4.2 by 2.5 to 3.5  $\mu$ .
5. *P. stephaniae* Zaleski, 300.  
Conidia 3 to 4 $\mu$ ; margin broad merging into dull green.
6. *P. palitans* Westling, 301.  
Conidia about 4 $\mu$ ; marginal zone bluish then green; traces of zonation at margin in age.

Xb. Conidia elliptical: color sometimes bright green.

7. *P. aurantio-virens* Biourge 210.  
Conidia 2.8 to 4.8 by 2.5 to 4 $\mu$ .
8. *P. janthogenum* Biourge no. 290.  
Conidia 3.8 to 5 $\mu$  by 2.4 to 4.2 $\mu$ , large spores, zones and streaks of color.

- XI. Colonies mostly in dull (even to dark) gray green or glaucous shades.....XII.

- XII. Zonation indistinct or reduced to ridges in the conidial mass; fascicles seen only at margin; conidial chains forming continuous crusts over the surface of mycelium.....*P. crustosum* Thom, 305.
- XII. Zonation usually evident especially in outer areas of colonies after growing for seven to ten days.....XIII.
- XIII. Zones narrow crowded (about 1 mm. intervals)..XIV.
- XIII. Zones fairly broad (about 2 mm. intervals).....XV.
- XIV. Fascicles visible only at margin.....XIVa and b.
- XIVa. Conidia subglobose to globose
1. Colonies shallow—300 to 400 $\mu$  deep; marginal zones bluish green.....*P. Blakesleei* Zaleski, 307.
  2. Colonies deeper—more or less floccose 800 to 1000 $\mu$  deep.....*P. porraceum* Biourge, 309.
- XIVb. Conidia elliptical; reverse orange; colonies rather deep often appearing floccose.....*P. aurantio-griseum* Dierckx, 310.
- XV. Zones fairly broad with fasciculation especially prominent in the marginal zones after one to two weeks of growth.....XVI
- XVI. Conidia becoming subglobose to globose when ripe.....*P. expansum* series XVII.
- XVI. Conidia persistently elliptical.....XVIII.
- XVII. *P. expansum* series; conidia about 3 to 3.4 $\mu$  in long axis.....XVII.
- XVIIa. Probably described from related strains.....*P. expansum*, Link, 315.  
*C. alphitopus* Secretan, 316.  
*P. crustaceum*. B. Coremium, 317. Fries.  
*Floccaria glauca* Greville, 317.  
*P. glaucum* Link (Wehmer), 319.  
*P. glaucum* Link (Sopp), 321.



- XVIIa. Probably described from related strains..... *P. leucopus* (Persoon) Biourge, 321.  
*P. malivorum* Ciferri, 322.  
*P. plumiferum* Demelius, 323.  
*P. variabile* Wehmer, 324.
- XVIII. Conidia elliptical.....XIX.
- XIXa. Differing from *P. expansum* by ellipticity of conidia and lack of characteristic odor.....XX.
- XIXb. Conidia at first almost oidio-form on citrus fruits.....XXI.
- XIXc. Colonies mostly in gray to gray green shades.....XXII
- XX. Conidia 3 to 4.5 by 2.5 to 3 $\mu$ ; floccose.....*P. aurantio griseum* Dierckx, 310.
- XX. Conidia 4 to 4.5 by 2.5 to 3 $\mu$ .....*P. elongatum* Dierckx, 325.
- XXIa. Conidia 4 to 4.5 by 2 to 3 $\mu$ ; giving cylindrical effects; with prostrate marginal coremia; sclerotia and perithecia described but not often encountered; on citrus fruits.....*P. italicum* Wehmer, 326.
- XXIb. Conidia 3 to 4.5 by 2.5 to 3.2; synonym of *P. italicum* (?).....*P. aeruginosum* Dierckx, 327.
- XXIc. Conidia 2.8 to 3 by 2.2 to 2.7 $\mu$ ; with the prostrate or ascending marginal fascicles of *P. italicum* but smaller conidia.....*P. ventuosum* Westling, 328.
- XXIIa. Conidia gray green to green; coremia sporadically developing in conidial areas; conidia 2.7 by 2.3 $\mu$ .....*P. juglandis* Weideman, 329.
- XXIIb. Conidia about 4 by 3 $\mu$ ; colonies olive green to gray with conidial areas running out in radiating lines at margin (stellate); reverse yellow to black brown.....*P. schneegii* Boas, 330.
- XXIIc. Conidia 2.8 to 3.2 by 2.2 to 2.5; conidiophore flexuous; odor peculiar; colonies gray green :..*P. urticae* Bainier, 331.
- XXIIId. Synonym.....*P. flexuosum* Dale, 332.

- XXIIe. Probably synonym.....*P. aeruginosum* De-  
melius, 333.
- XXIIIf. Conidia 3 to 3.5 or even 4 by 2.5 to 3 $\mu$ ; colonies  
gray to gnaphalium green; (close to preceding  
without odor).....*P. patulum* Bainier.  
334.
- XXIIg. Conidia 3.5 to 5 by 2.4 to 3 $\mu$ ; colonies bluish  
green to gray-olive to mouse gray in age.....*P. Polonicum* Zal.,  
335.
- XXIIh. Conidia up to 4 by 3 $\mu$ ; colonies dull dark green  
near artemisia green (Ridgway); reverse in  
orange red such as testaceous.....*P. brunneo-violaceum*  
Biourge, 336.

*Coremiella*

- XXV. Fasciculation fairly general throughout the  
colonies; fascicles or coremia crowded inter-  
spersed with single conidiophores but pre-  
dominating.....XXVI.
- XXVI. Conidia globose or subglobose.....XXVII.
- XXVI. Conidia mostly elliptical.....XXVIII.
- XXVIIa. Colonies blue green to dark olive gray, 300 to  
500 $\mu$  deep; reverse deep orange to brown;  
conidia mostly 3 $\mu$  or less.....*P. corymbiferum*  
Westling, 340.
- XXVIIb. Closely related.....*P. hirsutum* Dierckx,  
341.
- XXVIIc. Closely related.....*P. clavigerum* De-  
melius, 342.
- XXVIId. Closely related with taller coremia.....*P. divergens* Bain.  
and Sartory, 343.
- XXVIII. Conidia mostly elliptical.....XXIX.
- XXIXa. Yellow green; orange below; walls very granu-  
lar; conidia 3 to 3.5 by 2.5 to 3 $\mu$ .....*P. granulatum* Bain-  
ier, 344.
- XXIXb. Blue green; conidia 5 to 5.5 by 3.5 to 4 $\mu$ .....*P. granulatum* of Bi-  
ourge, 345.
- XXIXc. Possibly some members of this group.....*P. nigrescens* Jung-  
huhn, 346.

*Coremia*

- XXX. Colonies characterized by prominent coremia  
with simple conidiophores reduced or sup-  
pressed.....XXXI.

- XXXIa. Coremia large solitary upon apples, pears, etc... *P. expansum*, 315.
- XXXIb. Coremia large often several millimeters high on all media; conidia 4 to 4.6 by 3 to 3.3 $\mu$  in long chains forming in large heads..... *P. claviforme* Bainier, 350.
- XXXIc. Identical or nearly related..... *Coremium silvaticum* Wehmer, 350.
- XXXId. Coremia caespitose about 2 cm. in height; conidia about 6.5 by 2.2 to 2.8 $\mu$ ..... *P. Briardi* Vuillemin, 352.
- XXXIe. Coremia in culture forming broad areas with conidial mass supported by broad aggregates of conidiophores forming a pillar. Encountered in soil cultures and on insects..... *P. anisopliae* (Metch) Vuillemin, 353.
- XXXIf. Parasitic on cicadas; forming branching bushy coremia; conidia 5 to 6 rarely 7 $\mu$  by 1.5 to 2 $\mu$ ..... *P. cicadinum* von Höhnelt, 354.
- XXXIg. Colonies with erect or ascending columnar masses with Penicillium-like conidial masses scattered along this whole length..... *Isaria* sp., 355.

#### SUB-SECTIONS IN THE FASCICULATA

The Section Fasciculata includes a multitude of varieties or strains differing sufficiently in habit, color in conidial areas and color in the substratum to give individuality to parallel cultures. These differences are largely in shades of color or intensity of reactions whose determining factors are mostly unknown and whose stability through many generations of transfer upon artificial substrata is very doubtful. Nevertheless certain lines of division seem to be more or less stable and to separate series of strains either themselves cosmopolitan or widely represented by near relatives. Some of these forms have been maintained in culture for many years without loss of identity. Others less individual or less specialized leave the worker doubtful of the continuity of his own strain which he has sought to maintain pure.

As a working basis for separation we are ranging the species assigned to the section into sub-sections and separating these sub-sections into two groups: A—Sub-sections in which the aerial growth in Czapek's solution agar consists of a mixture of simple conidiophores with fascicles of conidiophores but with the simple conidiophores predominating and B—Two sub-sections characterized by fasciculate conidiophores and becoming practically coremia alone in the sixth. The second and fifth



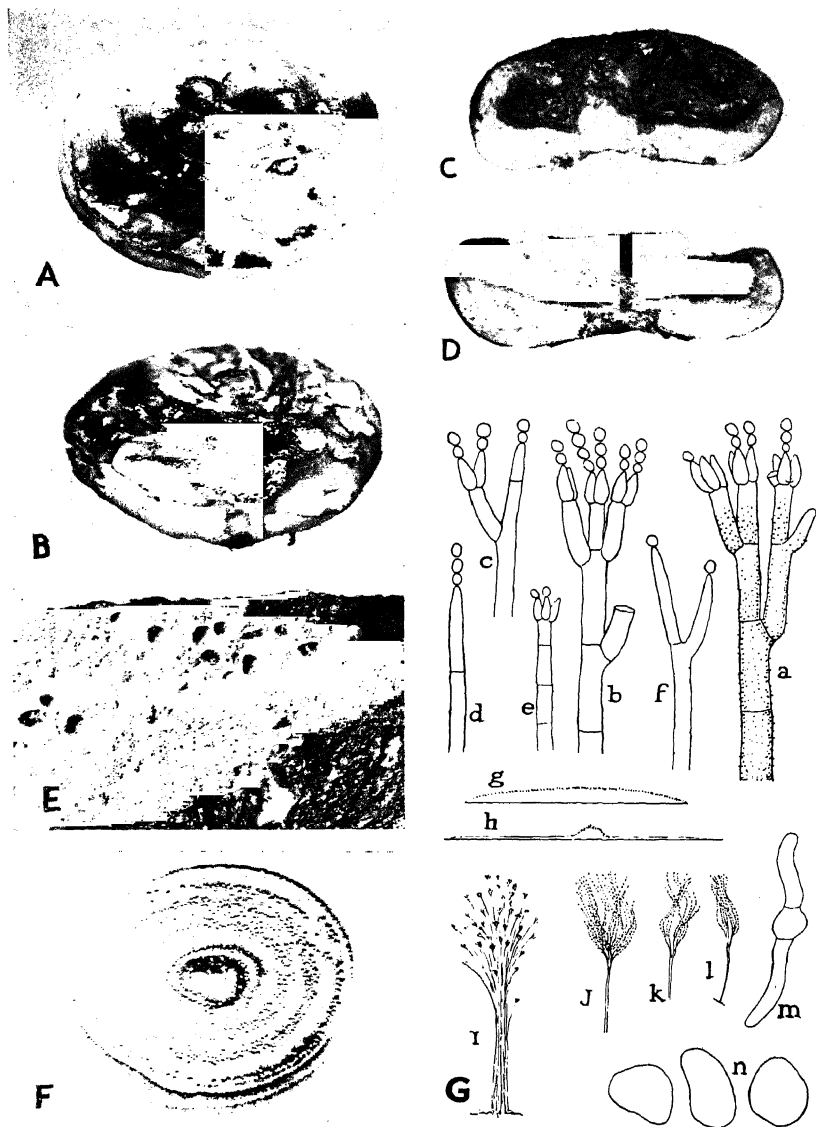


FIG. 58. *P. gladioli* after McCulloch and Thom: H, Detail of penicillus; habit sketches; F, zonate sclerotigenous culture; A, B, C, D, rotting *Gladiolus* conus; E, sclerotia magnified.

of these sub-sections contain species which might be interchangeable and necessarily will involve some confusion; the others are more readily separable.

- A. Sub-sections showing fascicles and simple conidiophores usually closely aggregated in culture; simple conidiophores usually predominating.
  - Sub-section 1. *Sclerotigena*: arbitrary segregation of sclerotium producing species.
  - Sub-section 2. *Aeruginosa*: conidial areas in strongly bluish green colors.
  - Sub-section 3. *Viridicata*: conidial areas in bright green to yellowish green shades.
  - Sub-section 4. *Glaucæ*: conidial areas in dull green or only transiently bluish green, then green or dull yellowish greens.
- B. *Coremia* predominating and characterizing the colony.
  - Sub-section 5. *Coremiella*: colonies in which fasciculation is fairly strongly marked throughout the colonies.
  - Sub-section 6. *Coremium*: colonies characterized by erect and usually separate coremia.

#### Fasciculata-Sclerotigena.

1. Sclerotia abundantly and regularly produced at 20–38°C.
  - a. fascicles of conidiophores, coremia and blue green conidia produced only at cooler temperatures —about 15°C., or lower. .... *P. gladioli*, 285.
2. Sclerotia or perithecia sporadically obtained from particular strains or combinations of strains or under conditions thus far undefined. .... *P. italicum*. No. 326

285. *P. gladioli* Machacek (Machacek, J. E.). Quebec Soc. for the Protection of Plants Ann. Rept. 19, (1926–27) pp. 77–86, (1927) 1928. Independently published under the same specific name by McCulloch and Thom, Science N. S. 67: no. 1730, p. 216–217, 1928; see also Jour. Agr. Res. 36, no. 3, pp. 217–224, Pl. I, 1928. Description follows McCulloch and Thom (see fig. 58).

Colonies presenting two aspects: When grown at temperatures above 20°C., predominantly consisting of sclerotia with few and inconspicuous conidiophores; when grown at 15°C. or lower, showing abundant green conidial areas with delayed or partially suppressed sclerotium formation. Upon Czapek's solution agar at 20° to 24°C. producing a thin aerial felt of mycelium with sclerotia beginning about the sixth day and developing in successive concentric zones, and giving the characteristic appearance of the species; sclerotia 140 $\mu$  to 540 $\mu$  in diameter, at first cream to light pinkish tan, in age very pale brown or tan, smooth, and composed of

thick-walled cells  $8\mu$  to  $12\mu$  in diameter, retaining their vitality several months; reverse light pinkish cinnamon; drops of orange-yellow fluid more or less conspicuous; odor none, conidiophores few, scattered and inconspicuous among the sclerotia, often very long (up to 2 mm.) and about  $2\mu$  to  $3.6\mu$  in diameter, later developing in more or less conspicuous tufts, fascicles, or complex branching coremia in the center of the colony and definitely green (bluish gray-green); penicillus consisting of the main axis of the conidiophore with or without one or two branches, bearing few metulae  $10\mu$  to  $12\mu$  long and verticils of few sterigmata  $12\mu$  to  $14\mu$  by  $1.5\mu$  to  $2\mu$  with tapering rather than acute points, and conidia elliptical-fusiform, smooth, hyaline,  $2.8\mu$  to  $3.6\mu$  by  $2.5\mu$  to  $3\mu$ , adhering in long chains in fluid mounts, more or less parallel then tangled as seen in the penicillus; swelling to  $6\mu$  or  $7\mu$  in diameter and germinating by one or two tubes.

Colonies grown at  $14^{\circ}$  to  $15^{\circ}\text{C}.$ , producing abundant mycelium and conidial areas gray-green (light dull glaucous blue, glaucous blue, and greenish glaucous blue of Ridgway (5)), to the very margin of the colony; sclerotium formation delayed and reduced, not dominating the growth; conidiophores partly simple, partly aggregated into coremia, tending to be longer and coarser, and when in coremia commonly  $3\mu$  to  $4.5\mu$  or even  $6\mu$  in diameter with walls pitted or roughened; penicillus coarser, more branched, and with larger verticils of sterigmata and the elements varying in diameter but following the conidiophore in being coarser than in the colonies grown at higher temperature; conidia not different from the conidia produced at higher temperature.

The conidial form described and figured here complies closely with the description of *Penicillium divergens* of Bainier and Sartory (1) and would have been identified as that species except for the entire lack of sclerotia in that species.

Found as a cause of decay in gladiolus corms. Specimens of natural and artificial infections on gladiolus corms and dry cultures have been deposited in the herbarium of the Office of Mycology and Disease Survey, Bureau of Plant Industry, United States Department of Agriculture.

In a culture contributed by Birkinshaw (5034.65) when grown upon slanted Czapek agar in the incubator at  $15$  to  $20^{\circ}\text{C}.$ , the drops were deep orange red or vinaceous rather than orange yellow as seen at room temperatures, no sclerotia appeared in twelve days and the general appearance of the colony was close to that of *P. corymbiferum* Westling.

This species appears to have been isolated during the same year by

J. E. Machacek at Macdonald College, Quebec, O. H. Elmer at Manhattan, Kansas and Miss McCulloch at Washington. Machacek presented a paper at the meeting of the Quebec Society for the Protection of Plants at Macdonald College, March 30, 1927; after this he mailed his culture without information as to his announcement of it as new, to Thom for identification so that it was received May 4, 1927 and he was advised of its isolation and description by us. He subsequently acknowledged that his paper had been presented at a meeting and the date of such presentation, but withheld the name and description used. Every effort to obtain information failed until the report containing his paper was received by the U. S. Department of Agriculture Library, April 3, 1928. In the intervening period both of Miss McCulloch's Papers were published. There is no question as to identity of the organisms. We have, therefore, substituted our own description of the species but ascribed priority of publication to Machacek. Professor Elmer's courtesy in this matter is hereby acknowledged.

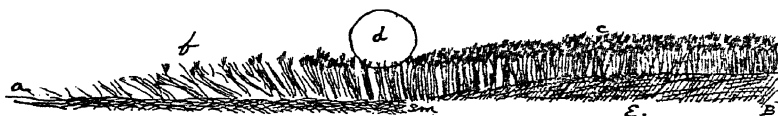


FIG. 59. The *P. cyclopium* type of colony with fasciculation evident only at margin: Diagrammatic radial section (magnified 25 times); *ab*, agar line; *c*, dense conidial layer; *d*, large exuded drop; *e*, elevated or bulged area of substratum and proliferated mycelium; *f*, fasciculation of conidiophores evident in the marginal 3 mm.

*Sub-section 3. Aeruginosa.* Colonies in blue green shades, with fasciculation evident in the white marginal area often obscured in the older areas and even at the margin in old cultures (fig. 59).

The species assigned to the blue green group have been roughly separated into three series based upon zonation.

- (a) The *P. cyclopium* series which are hemizonate according to Biourge, *i.e.*, zonation when it appears at all, appears late in colony development and remains usually indistinct..... See no. 286.
- (b) The *P. martensii* series with well marked zonation usually rather broad, usually yellow turning to maroon in reverse. .... See no. 291.
- (c) The *P. Johannioli* series with zones narrow and crowded at the margin; yellow in reverse reduced or suppressed followed by purplish or almost black shades in the central area..... See no. 293.



Many strains in each of these three series are encountered hence the identity of a strain found with one described is not usual although it happens often enough to convince the worker that fairly permanent types do exist. These described forms constitute therefore centers of what may be called "series" or aggregates of strains with common morphology and general agreement in physiological activity but differing in the shade of color in conidia and substratum and in the quantities of mycelium and spores produced.

286. *P. cyclopium* Westling. Arkiv för Botanik 11, pp. 55-56, 90-92, fig. 15, 57. 1911.

Colonies upon prune gelatine azonate, with a granular (tufted or coremiform—C. T.) appearance noticeable to the very center, slightly floccose, spreading broadly with conidial area rather slowly sky blue (C.d.C. 367, 362, 363) which merges imperceptibly into a broad white margin, then becoming green (338, 335, 330), finally greenish gray or even dark brown, gelatine very slowly and only partly liquefied; reverse yellow to orange; hyphae 2 to 5 $\mu$ , occasionally 8 $\mu$  in diameter, with most commonly sphaerocrystals of calcium oxalate scattered among them; odor none recorded; conidiophores arising from aerial hyphae, most frequently singly, but appearing as numerous tufts, fascicles or coremia, longer especially in the older central area from which the colony gradually thins out toward the margin, with walls mostly very finely "warty," 90 to 750 $\mu$  or longer by 3 to 5 or even 6 $\mu$ , penicillus 45 to 120 $\mu$  long, figured as main stalk and rather long appressed branch or branches carrying the verticils of metulae, but with branching absent in the smaller fruiting masses; metulae only slightly roughened, 9.5 to 14 by 3.2 to 4.4 $\mu$ ; sterigmata 8 to 9 by 2.6 to 2.8 $\mu$ ; conidia smooth, globose, 2.6 to 3.2 $\mu$ ; swelling in germination to 6 to 7.5 $\mu$ .

Species found on rotten fruit of *Actaea spicata*, *Fragaria moschata* and upon roots of *Uragaga ipecacuanhae* Bail. in Norway, showing resemblances to *P. corymbiferum* West. Colonies grew poorly at 30° to 31°C. and changed blue litmus gelatin to red, and grew well upon malt-extract gelatin, on plum agar, potato, bread, maranta starch, sugar solution and milk in which the mycelial margin of the colonies was yellow.

A culture received from Westling as this species proved to be incorrectly labeled. Another received through Miss Dale proved also unsatisfactory. Biourge's organism however seemed to correspond fairly well with the description. Our notes upon it have been combined with Biourge's (see Biourge, Monogr. La Cellule 33: fasc. 1, pp. 138-139; Col.

Pl. XI, Cart 382; Pl. XIX, fig. 114, 1923): Colonies in Czapek's solution agar included in zonate group by Biourge, zonate only in age in our cultures, velvety, thin at the margin rising to about  $500\mu$  in depth at the center with a tendency to overgrowth of white mycelium in center, with conidiophores crowded, largely in fascicles or tufts without definite coremia, with marginal area during the growing period white passing slowly into bluish gray green; reverse pale yellowish becoming purplish in center, or greenish in irregular areas; odor noticeable, difficult to characterize; measurements as given by Biourge correspond fairly well with Westling; conidiophores with walls rough,  $3\mu$  in diameter; penicillus about  $60\mu$  long, figured as a main stalk with partly diverging branch, each bearing verticils of metulae; branches in pairs 25 to 40 by  $2.5$  to  $3\mu$ ; metulae 9 to 13 by 2 to  $3\mu$  in threes or fours, with apex swollen, almost vesicle-like; sterigmata 7 to 8 by 2 to  $3\mu$ , at length rough, in verticils of 2 to 7; conidia globose  $3\mu$  in diameter, described as subglobose or  $3.5$  to  $3.8$  by  $3\mu$  slightly roughened; in our cultures  $4\mu$  and smooth.

Biourge's no. 382 (our no. 4733.48) as received appears to lack zonation only in comparison with his description; the colony description given was therefore taken from our own data.

Numerous strains with the morphology of *P. cyclopium* as represented by Biourge's culture and description have been studied. These include three from honey bees contributed by C. E. Burnside and several from tulip bulbs, in which they were reported as attacking and destroying the growing point.

A culture contributed by Biourge as *P. corymbiferum* Westling proved to be nearly related to *P. cyclopium* but with zonation of the colonies more pronounced, colonies a little deeper in mass, and conidia somewhat larger. Our notes upon this organism follow: Colonies azonate at first, indefinitely zonate at the margin in old and drying cultures, plane in shallow layers of nutrient, buckled and broadly radiately wrinkled in deep agar, with margin broad white, with granular fasciculate bluish green zone of developing conidia and a central area darker than bluish green, the whole felt of mycelium and conidiophores perhaps 700 to  $1000\mu$  deep in deep agar, shallower in thin areas; reverse transiently yellow-pale passing to pinkish buff fading to a pale shade of the same or cinnamon buff in age; drops colorless in deep agar; odor definite moldy; conidiophores 3 to  $4\mu$ , all walls rough; penicillus one-sided verticil of central axis plus one or more branches often unequal more or less closely clustered; metulae 12 to  $15\mu$  by  $3\mu$ ; sterigmata about 10 to  $12\mu$  by  $2\mu$  tending to tapering apex; conidia about  $3.5\mu$  with suggestion of roughness.

Culture no. 4733.43, *P. corymbiferum* fide Biourge, thus gives certain prominent characters: Zonation, as ridges in a deep dense colony; fasciculation prominent in three marginal zones; fairly deep, probably 600 to 900 $\mu$ ; marginal zone white broad 2 to 3 mm. in slant, next two zones with bluish then deep blue green zones 4 to 5 mm., in width; reverse in a pale salmon tint; odor, indistinct.

5. *P. flavo-glaucum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 130-132; Col. Pl. I, Cart. 73; Pl. II, fig. 10. 1924.

Biourge's description has been combined with our own study of his culture: Colonies on wort gelatine, velvety or nearly so, spreading broadly as a growth 200 to 400 $\mu$  in depth. Zonate in the outer areas not at center, pale to dull green with only a transient marginal bluish green in some cultures, described as producing sessile coremia in older areas upon some media, but found in certain transfers to produce tuberculate masses or crusts up to 500 $\mu$  or more in depth, consisting of closely packed parallel chains of conidia; with white marginal zone about 500 $\mu$  broad, but showing radiating and ascending hyphae carrying conidia production almost to the very edge of the colony; reverse pale yellow or cream; conidiophores rough, pitted, 4.5 to 5.5 $\mu$  (even 6 $\mu$  in our transfers) in diameter; penicillus with walls rough, about 50 $\mu$  long, figured as main stalk and a rather long appressed branch or branches from different nodes, producing metulae and sterigmata often at 2 or more levels; branches in twos, 22 to 24 by 3.3 $\mu$  granulate; metulae reported as 7.5 to 12 by 2.2 to 2.8 $\mu$ , in twos or threes in ours, usually, one at each septum, primary up to 30 $\mu$  long, secondary when present about 20 $\mu$  long; sterigmata 10 to 14 to 16 by 3 $\mu$  in twos or threes (few to each verticil); conidia described and figured as elliptical 4.5 to 5 by 2.8 to 3 $\mu$ , in our cultures from Biourge's no. 73, subglobose mostly 4 $\mu$  less commonly 5 $\mu$ , with occasional elliptical pyriform or double sized conidia.

Biourge's type no. 73 (our no. 4733.61a) was received in 1924.

289. *P. conditaneum* Westling. Arkiv för Botanik 11: 52, 63-65, figs. 46, and 2. 1911.

Colonies in prune gelatine, dark green (C.d.C. 329, 330, 334, 339), bluish green at first on malt extract gelatine and on potato, gray green to gray black in age, with a narrow white sterile margin, (resemblance to *P. solitum* is indicated); reverse bright yellow or at times colorless; gelatine slowly and partly liquefied; odor slight; vegetative hyphae coarse 3 to 6 or even 8 $\mu$  in diameter, with abundant vacuoles and frequent

anastomoses; conidiophores arising from submerged or creeping hyphae, very short to very long up to  $800\mu$  by 4 to  $6\mu$ , with walls, smooth, uneven or warty, and with penicillus 55 to  $160\mu$  long, consisting of a branch or verticil, metulae, and sterigmata; metulae clavate 12 to 18 by 4 to  $6\mu$ , or wanting and replaced by long sterigmata; sterigmata 8 to 9.6 by 3 to 3.4 or even  $4\mu$ ; conidia pyriform to globose smooth, commonly 4 to  $4.6\mu$ , ranging from 3.6 to  $5.6\mu$ , and swelling in germination to 7 to  $9.6\mu$ , in chains which break up readily; perithecia and sclerotia not found.

Species found on *Ribes nigra*, in association with *P. majusculum* Westling, related in appearance to *P. solitum* Westling, but darker in color and more uniform in spore size. Westling reports sphaerocrystals and columns of calcium oxalate among the hyphae. Colonies grew well on all media tested.

Colonies sporulate up to 30 to 31° and produce mycelium up to 35°C. Type culture received from Westling and studied but lost, no. 2538.

We are not satisfied to attach the name to any organism now in our collection.

Turesson (1911) reported this species as killing honey bees when fed in quantity.

Biourge (Monogr. La Cellule 33: fasc. 1, p. 143) received *P. frequentans* Westling from Amsterdam labeled *P. conditaneum*, hence his note is a summary drawn from Westling's paper; he places it in the zonate group.

No. 4777.18 received from Pribram as *P. convitaneum* Westling was not this species as described by Westling although the name was apparently a corruption of this name as proposed by Westling.

3. *P. janthogenum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 143-145; Col. Pl. II, Cart. 82; Pl. III, fig. 13. 1923.

Colonies on wort agar at times zonate, bluish green, then green (C.d.C. 347-348), with true coremia on potato, bread, and rarely on bean agar; reverse violaceous and yellow, sometimes red and yellow; drops none; odor woody; gelatine liquefied, uncolored, then pale brown, acid; conidiophores about  $4\mu$  in diameter, walls all smooth; penicillus about  $50\mu$  long or  $35\mu$  without branches, figured as main stalk and one long, septate appressed branch bearing metulae and sterigmata at slightly different levels but making a compact mass; branches 25 to 35 by  $4\mu$  in pairs, or wanting; metulae 16 to 20 by 3 to  $3.5\mu$ , in threes or fours, with apex enlarged, vesicle-like; sterigmata 12 to 14 even to 20 by 3 to  $4\mu$ , in verticils of 2 to 4; conidia ovate 4.8 to 5 by 2.4 to  $4.2\mu$ .

Biourge's no. 82 (our no. 4733.77) on Czapek's solution agar produced colonies appearing zonate and fasciculate in the marginal areas only, becoming densely felted within and up to 1 to 3 mm. deep, hence frequently reported as floccose in routine records, at first white then shades of gray green, and blue green; reverse variously colorless, with citrine zones, becoming reddish brown in spots streaks or zones under various conditions of culture; drops colorless, numerous in the outer 2 to 3 zones; odor absent at first, becoming fairly strong in two weeks; conidiophores varying in length, about 3 to  $4\mu$  in diameter, with walls smooth or possibly showing traces of pitting; penicilli consisting the main axis and appressed branches up to  $20\mu$  long, metulae 10 to  $15\mu$  long, several in the verticil, and sterigmata about  $12\mu$  long; conidia elliptical about 4 by  $3\mu$ .

291. *P. martensii* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 152-154; Col. Pl. II, Cart. 118; Pl. III, fig. 14. 1923.

Colonies in wort gelatine, zonate, blue to blue green, to dark shades in age, with broad white marginal zone, reverse in yellow shades mostly, gelatine liquefied and yellow; odor weak, or none; conidiophore about  $3\mu$  in diameter; penicillus about  $40\mu$  long, with all walls smooth, figured as main stalk with or without a fairly long appressed primary branch and with or without verticils of short secondary branches producing the metulae and sterigmata at a fairly uniform level; branches, primary about  $30\mu$  long, secondary 6 to  $8\mu$ , both 2.5 to  $2.8\mu$  in diameter; metulae varying in length from 6 to  $14\mu$  by 2.5 to  $3\mu$ , in twos or threes; sterigmata 10 to 14 by 2.5 to  $3\mu$ , in groups of two to five; conidia 3.4 to 4 by 2.4 to  $3\mu$ , persistently elliptical.

Colonies of Biourge's no. 118 (our no. 4733.87) grown upon Czapek's solution agar, zonate, with a felted aerial mycelium below an apparently velvety growth, with outermost zone 1 mm. broad, submerged, a transition zone 2 mm. broad, white, then pale blue to rich blue green conidial zones, with conidiophores partly fasciculate, tufted, almost coremiform, with some overgrowth in center; reverse and agar yellow at first then rapidly changing to deep reddish brown through the whole mass of medium; odor faintly fragrant; conidiophore about  $4\mu$  in diameter with walls delicately pitted; penicillus as described by Biourge, except that the number of sterigmata is usually greater, and conidia 3.5 to 4 by 3 to  $3.5\mu$  or subglobose 3 to  $3.5\mu$ ; inoculated into apples, this species produced no rot.

The contrast between the grass green (*viridis*, *prasinus*) and the blue-

green (aeruginous) members of the zonate fasciculate section is superficially conspicuous as typical strains are met in culture. Microscopically they furnish little morphological basis for separation. Changed from substratum to substratum the range of colors in any member of the series may broaden sufficiently to overlap the reactions of other strains originally regarded as easily separated. Biourge fixed upon the strain described as *P. martensii* as a distinct species. We are accepting it as an outstanding individual strain in a group ranging toward the bright greens of *P. viridicatum* on the one hand and the glaucous green of *P. expansum* on the other.

The different strains assigned here vary considerably in details of growth habit; in the rapidity and intensity of color and color changes in the substratum. Some of them retain the ellipticity of the conidia throughout; others show most of the conidia almost subglobose when ripe (no. 4975.131).

There are many aeruginous strains with conidial areas giving the broadly zonate appearance after the first few days of growth but in reverse yellow to orange rather than maroon to deep orange brown. Among them are several from Logan, Utah, isolated by C. M. Tompkins from decaying sugar beets. Until more careful quantitative means of separation are found these may well be "lumped" in *P. martensii*.

Similarly another series of strains differing but little in morphology lack, or only show traces of yellow or orange in the mycelium but develop dark purplish shades in reverse in older colonies. Zaleski's *P. Johannioli* has the general structure and reactions cited, hence may stand as a composite species for such materials (see no. 293).

292. *P. majusculum* Westling. Arkiv för Botanik 11: 51-52, 60-62, figs. 1 and 45. 1911.

Colonies in prune gelatin, at first blue green (C.d.C. 362, 363), then dark green (C.d.C. 334 and related), becoming brown in age (one month or more); not floccose, with narrow sterile margin; reverse pallid yellow; gelatine slowly and partly liquefied; odor weak, moldy; conidiophores arising from creeping hyphae, smooth at first then commonly slightly verruculose, up to  $550\mu$ , commonly 150 to  $300\mu$  by 4 to  $6.5\mu$ , with penicillus 45 to  $195\mu$ , commonly 90 to  $150\mu$  long, with or without branches preceding the formation of verticils of metulae; metulae 12 to 20 by 4 to  $6.5\mu$ , occasionally 1-septate, hence 2-celled; sterigmata 10.5 to 15 by 3 to  $3.6\mu$ ; conidia at first elliptical then oval-globose to globose, smooth, 4.5 to  $6\mu$ , swelling in germination to 7.5 to  $9.6\mu$ . Sclerotia or perithecia not reported.

The culture received from Westling as his type (our no. 2542) never gave colonies agreeing very well with his general description and produced conidia averaging considerably smaller than Westling's measurements. There may be therefore some doubt as to its validity as type but it agrees in so many points with his description that the name is retained. Our notes follow: Colonies upon Czapek's solution agar, slowly and rather restrictedly growing, zonate, and appearing fasciculate under the handlens, forming a shallow growth about  $200\mu$  deep, blue green in the outer 10 to 15 mm., fading to light and dark gray zones within; reverse variously uncolored and vinaceous to drab shades, in different cultures; odor strong "moldy" suggesting *Actinomyces*; conidiophores mostly very short, up to  $200\mu$  by 4 to  $4.5\mu$ , with walls pitted or rough; penicilli consisting of main axis and 1 or 2 appressed branches up to  $40\mu$  long, bearing metulae varying from the usual type up to  $40\mu$  long and 1-septate as metulae take the place of branches in the system, with conidia produced in tangled chains; sterigmata commonly 12 to  $14\mu$  long, others up to 18 or  $20\mu$ , varying with the culture and the position in the penicillus; conidia at first elliptical about 4 by 1.5 to 2 becoming variously  $4\mu$ ,  $4.5$  by  $4\mu$ , or up to 6 by 4 to  $5\mu$ , but mostly subglobose  $4\mu$  or  $4.5$  by  $4\mu$ .

Biourge (in Monogr. La Cellule 33: fasc. 1, pp. 137-138; Col. Pl. IV, Cart. 370; pl. VI), gives: Colonies in Raulin or wort gelatine, in the zonate (Hemizonate or faintly zonate in age in our cultures) group, blue green, becoming violaceous brown in age, velvety, with a narrow white marginal zone during the growing period; with occasional cells 14 by  $11\mu$  whose nature was not determined; reverse a greenish yellow; gelatine slowly liquefied.

In our cultures, Biourge's organism (our no. 4733.86) gave colonies upon Czapek's solution agar almost velvety when young appearing fasciculate 200 to 250 in depth, with center buckled, gray green shading to mouse gray in age; in reverse with center dark, remainder colorless with drops pink to lavender in color and drying to leave a lavender residue; penicillus 35 to  $40\mu$  long, figured as a verticil of metulae with 1 metula bearing a secondary verticil, the others bearing sterigmata; conidiophore 4 to  $5\mu$  in diameter, with walls delicately granular; metulae 14 to 18 by 4 to  $5\mu$ , commonly in 4's sometimes with secondary short verticils; sterigmata 10 to 16 by 3 to  $4\mu$  in groups of 2 to 4; conidia 2.5 to 6 by 2.5 to  $5\mu$ , commonly 6 by  $4\mu$ , in our cultures globose 3 to  $5\mu$  in diameter.

Biourge's no. 370 (our no. 4733.86) was figured and described as *P. majusculum*, but appears to have been unsatisfactory to him; he refers to its relationship as possibly *P. brevi-compactum*. His culture appears to be near to if not identical with our no. 2542, which was sent to him.

293. *P. Johannioli* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 453-454; Taf. 40; Zaleski no. 402.

Colonies in neutral Raulin with 10 per cent gelatine quickly growing becoming 4.5 to 5 cm. in diameter in twelve days, liquefying the gelatine very rapidly beginning about the tenth day, thin, velvety or with some funiculose masses or ropes of aerial hyphae, either entirely plane or slightly plicate, definitely zonate throughout with zones narrow (dense?) with an elevated point or umbo in center, with an overgrowth of scattered delicate gray aerial hyphae; margin during the growing period 2 to 3 mm. wide; in color blue green when young, C.d.C. 363 to 358, 362, and later 339, 340, 345; reverse showing crowded zones, plane or with a few radiate but delicate wrinkles, with the mycelium giving a typically radiate appearance; in color pale yellow 171 to orange about 108; liquefied gelatine in yellow or orange color; drops uncolored, few; odor weak; conidiophores 400 to 500 $\mu$  by 3 to 4 $\mu$ , straight, simple or occasionally branched; penicillus mostly 40 to 55 $\mu$  long; branches (rami) 18 to 32 by 3 to 4 $\mu$ , in number 2 or 3, with walls smooth or slightly roughened; metulae 10 to 13 by 2.5 to 3 $\mu$ , in groups of 4 to 6, commonly vesicle-like at the apex, figured as uneven in length; sterigmata about 9 to 10 by 2 to 2.2 $\mu$ , commonly 6 to 8 in the verticil, straight or slightly incurved; conidia 2.5 to 4 by 2.2 to 3.5 $\mu$ , mostly subglobose, some ovate, smooth, not persistently adherent in chains or masses.

Habitat: Species isolated from rotting onion bulbs in Poznan, Poland.

Biourge regarded this as a polyverticillate form of the series containing *P. roseo-citreum* Biourge, *P. notatum* Westling and *P. brunneorubrum* Dierckx. Zaleski therefore puts it among the Radiata. Our notes follow: Type strain growing more rapidly at about 20°C., than at 30°C. or above; colonies on Czapek's solution agar at 20°C., spreading slowly (25 mm. in diameter in seven days) buckled in center with a few wrinkles radiating toward the margin, at first azonate then slowly producing narrow or crowded marginal zones, with conidiophores definitely seen in fascicles in the marginal areas or zones, producing a total mass up to 500 to 800 $\mu$  deep, with conidial areas deep blue green (C.d.C. 362, 363), and later shades of green; reverse at first in yellowish shades, later showing dark or purplish black colors; drops, large, colorless; odor?; conidiophores 100 to 200 $\mu$  long; penicillus 30 to 35 $\mu$  long, with primary branches up to 30 $\mu$  long, sometimes secondary branches up to 15 $\mu$  long, metulae about 12 $\mu$  long sometimes with sterigmata and secondary metulae in the same verticil; chains of conidia in parallel more or less adherent and slightly diverging groups verticil by verticil; sterigmata about 10 $\mu$



long; conidia mostly elliptical 4 by 2.5 to 4 by 3 $\mu$  or less often subglobose 3.5 to 4 $\mu$ .

Culture no. 5010.11 received from Baarn in July, 1928, is apparently type.

In studying a single series of species from decaying sugar beets we find a form no. 4975.238 which apparently belongs with *P. johannioli* and several strains whose divergence is not great enough to make more than varietal differences hence until some one studies this series intensively they may well be kept together.

*Sub-section 3. Viridicata* or, the *P. viridicatum* series. Colonies with fascicles showing in the marginal area only, coremia rare and then under ill defined conditions in very old culture; more or less definitely zonate, especially in the marginal areas during the latter part of the rapidly growing period; green between viridis and prasinus of Saccardo's *Chromotaxia*, with only transient bluish in the youngest conidial zones and in many cultures purple brown in age; conidiophore walls pitted or granular; hyphae mostly coarse: includes nos. 296 to 301.

Among bright green species placed elsewhere *P. psittacinum* Thom no. 270, might from its color and habit be placed here. Careful study of its colonies shows that ropes of hyphae predominate rather than fascicles.

Westling, in 1913, described *P. viridicatum* sending us his culture which became no. 2552 in our collection. Study of this culture quickly lead to recognition of the existence not of a single well marked species typified by Westling's culture, but of a whole series of forms with so much in common as to justify grouping them together. In this series, colonies upon Czapek's solution agar are zonate (at least at the margin in age), at times showing a bluish tinge at the very margin, changing quickly to bright green shades near Saccardo's viridis and prasinus cited by Westling as 337, 338, 347, 348 in Klincksieck and Valette's "Code de Couleurs," then becoming more or less rapidly rich brown in age, occasionally with sterile yellow areas among the green conidial areas; in reverse varying from colorless with a tinge of yellow to bright yellow, passing in some to orange or reddish orange, in some to brick red or maroon; marginal white areas are broad, show fasciculation of hyphae which is commonly obliterated by crowding and thickening of the conidial masses in the older zones; conidiophores are rough, show rather complex branching in the penicillus and produce conidia from subglobose to globose, varying in long axis in the different strains from 2.8 to 3.5, 4 $\mu$  or even larger. The following list shows something of the source and distribution of strains identified as belonging with this series.

Many more of these forms have been seen: 2552 from Sweden—Westling; 3028 from Kansas—Thom; 47 from Connecticut—Thom; E5454 from Connecticut soil—Esten; 2736 from England—Miss Dale; 4676PVI from Toronto—Miss Derick; 2643 from England—Miss Dale; 4613H from England—F. T. Brooks; 4601B1 from Maine, on *Vaccinium*—Stevens; 4658.37.7 from Cape Town, S. Africa—Putterill; 4933 from Heald, Pullman, Washington; 4270 from China—Dr. Chung; 4725.856 from Jamaica—C. G. Hansford.

296. *P. musae* Weidemann. Centralb. f. Bakt., etc., 2 Abt. 19: 687–689, fig. 3. 1907.

Colonies on grape-sugar gelatine, quickly growing, white on the second day, with colored conidial areas on the third, yellowish green, not blue green in all cases, with a broad white marginal zone, with conidial areas turning from green to brown in a few days; coremia produced especially at the margin upon bread, but clusters or fascicles suggestive of coremia appear among the crowded conidiophores in many cultures, with vegetative hyphae  $2.5\mu$  in diameter; gelatine quickly liquefied; conidiophores 3 to  $3.2\mu$  in diameter; penicillus consisting of main stalk 1 or 3 branches at the same or different levels, often slightly diverging and unequal in length, bearing the verticils of metulae; metulae in length about 6 times the diameter; sterigmata 5 to 9 in the verticil, conidia 2.2 to 2.8 by 2 to  $2.3\mu$ , elliptical or subglobose, swelling in germination to 4 to  $5\mu$  in diameter, produced in so great numbers as to make observation of the structure of the penicillus difficult.

Species found as yellow brown to olive brown patches on a banana.

297. *P. olivino-viride* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 132–133; Col. Pl. II, Cart. 22; Pl. II, fig. 12. 1923.

In series with *P. viridicatum* Westling or perhaps var. (?).

Colonies upon Raulin and wort gelatine, more or less definitely zonate according to the amount and richness of the media, velvety with more or less aerial network 400 to  $600\mu$  in depth, and with conidiophores more or less aggregated into tufts more conspicuous in the newer zones, which become definite coremia upon some media (potato), conidial area transiently bluish green then green (C.d.C. 328 and related shades), with broad white marginal zone in young colonies; reverse and agar colorless then pale yellowish to yellow and finally reddish brown (Ridgway; madder brown); drops yellowish; odor strong objectionable; taste intolerable; (no odor in our cultures—C. T.); conidiophore 2.5 to  $3.5\mu$  in

diameter, walls smooth, penicillus 40 to  $55\mu$  long, figured as a main stalk and rather long, slightly diverging branch bearing metulae and sterigmata at nearly a single level; branches in pairs, 15 to 30 by 2.5 to  $3.5\mu$ ; metulae 11 to 15 by 1.5 to 2.5, with apex commonly swollen, in twos or threes; sterigmata 10 by 2.5 to  $3\mu$ , in verticils of 2 to 5; conidia globose 2.5 to  $3.5\mu$ .

Biourge gives his culture no. 22 (our no. 4733.93) as *Penicillium* no. 29 of Miss Dale (Ann. Mycol. X, p. 464, 1912) and that it had been determined by Thom as *P. viridicatum* Westling, a species not discussed in full by Biourge and apparently unknown to him since at least four closely related strains have received separate names in Biourge's monograph.

298. *P. viridicatum* Westling. Arkiv för Botanik 11: 53, 88-90, fig. 14, fig. 56. 1911.

Colonies in prune-gelatine, with only slight floccosity, with margin broad, white, showing tufts, or green (C.d.C. 337, 333, 338, 309) coremium-like aggregations of hyphae or conidiophores; reverse yellow, to bright yellow, gelatine liquefied, between the sixth and twelfth days, with acid reaction; odor weak, drops of fluid prominent and abundant upon the growing mycelium; conidiophores arising from creeping hyphae, with walls mostly smooth, varying from 50 to  $600\mu$  in length and 4 to  $6.5\mu$  in diameter, and more or less combined, especially at the margin into small prostrate columns or coremia, penicillus 60 to  $180\mu$  long, figured as consisting of (1) a branch and main axis unequal in length, but usually forming long internodes, (2) verticils of metulae, (3) sterigmata and conidial chains; metulae 10.5 to 12 by 4 to  $5.6\mu$ ; sterigmata 8 to 9.6 by 3.2 to  $3.4\mu$ ; conidia smooth, globose, 3 to  $3.8\mu$ .

Species found upon roots of *Alkanna*, and branches of *alnus* in the pharmacy. Cultures grew well at  $30^{\circ}$  to  $31^{\circ}\text{C.}$ , changed blue-litmus-gelatine to red, and grew well upon malt-extract gelatine, upon sugar solution which became yellow, upon potato, upon bread, and upon milk in which the margin was yellow but the milk remained colorless. Westling's culture no. 2552 confirmed his description fairly satisfactorily; reverse was recorded as yellow, no reference was made to the change to reddish brown found in our cultures and his colony was recorded as persistently green, no report of change to brown!

*P. "viridiacum"* Westling appears in Wehmer, Mycol. Centralb. 2: p. 201, 1913, as a misprint.

299. *P. verrucosum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88.  
1901. See Biourge, Monogr. La Cellule 33: pp. 123-126; Col.  
Pl. I, Cart. 44; Pl. II, fig. 7. 1923.

Biourge gives: Colonies on wort gelatine zonate with conidial areas in shades of green (C.d.C. 333, 338, 343) in some media showing transient bluish green in the newest zones, with a broad white marginal zone, and with coremia-like tufts evident in the newer zones obliterated in the denser central area which becomes 800 to 1000 $\mu$  in depth and is again overgrown with white mycelium toward the center, or as white masses or warts; reverse at first yellow to orange, after several days showing a band of reddish brown, and dirty olive shades; odor of moldy potatoes; drops pale yellow; conidiophore 3 to 4 $\mu$  in diameter, in our cultures 4 $\mu$ , with walls rough pitted, penicillus figured as coarse, main stalk and branch each bearing metulae with occasional proliferations from metulae to produce secondary verticils with sterigmata at different levels; branches 15 to 18 by 2.8 to 4 $\mu$ ; metulae 10 to 12 even to 16 $\mu$  by 3 to 4 $\mu$ , apex broad vesicle-like; sterigmata 9 to 10 by 3 to 3.5 $\mu$ , in verticils of 2 to 4 (in our cultures about 7 $\mu$  long); conidia 2.8 to 4.2 by 2.5 to 3.5 $\mu$ , elliptical (our colonies show 2.5 to 3 by 3 to 3.5 $\mu$  or subglobose 3 to 3.5 $\mu$ —C. T.).

Biourge's culture no. 44 (our no. 4733.125), satisfies the description fairly well and belongs in the series with *P. viridicatum* Westling.

300. *P. stephaniae* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 451, 452; Taf. 40; Zaleski no. 1054b.

Colonies on neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 2.5 to 2.8 cm. in diameter in twelve days, with liquefaction of gelatine beginning at the fourteenth day and proceeding rapidly, outer areas rather thin, velvety, buckled giving the central area either cushion-like elevated or depressed (concave), indefinitely zonate (parum distincte), with all or more commonly the outer areas radiate wrinkled; margin (fimbria) 1 to 2 mm. in the growing period; in color white for the first six days becoming green shades such as C.d.C. 346, 347, 342, 343, 348 with ripening conidia and very dark shades of orange brown such as 173, 174 in old cultures; reverse at first in yellows 166, 157 later becoming orange 127, 128, 129, 130, and related shades; drops uncolored, small, abundant in the outer areas, less frequent in the central area; odor weak, suggesting rotting potatoes; conidiophores about 300 to 500 $\mu$  by 3.5 to 4 $\mu$ , straight or somewhat flexuous, with apices vesicle-like; penicillus 40 to 50 (or even 60 $\mu$ ) long, with walls

smooth, figured as with branches uneven in length and sterigmata borne at various levels; branches straight or incurved about 20 to 32 by 3 to 3.5, in groups of 2 to 3; metulae about 12 to 14 by 3 to 3.5 $\mu$ , in groups of about 5 to 7, clavate or with apices vesicle-like; sterigmata 10 to 11 by 2.2 to 2.5 $\mu$ , in verticils of about 6 to 10; conidia smooth globose or subglobose 2.5 to 3 $\mu$ , persisting in masses.

Habitat: Species isolated from sandy soil under pine trees, at 8 cm. depth, in "Długa Goslina" near Poznan in Poland.

Zaleski classes it among the "Hemiconcentrica 2° Inflata."

Zaleski's culture as received (our no. 5010.35) appeared to be heavily contaminated with some organism which transformed the growth into a sodden mass upon which a single spot of bright green finally appeared. Transfers from this spot gave colonies which correspond clearly enough to Zaleski's description to support the belief in identity. Our notes follow: Colonies upon Czapek's solution agar forming a mass 400 to 600 $\mu$  deep in the deeper areas, fasciculate, narrowly zonate with zones more distinct in the outer area than toward the center, with outer 2 or 3 zones more or less submerged, about two more zones uncolored and the inner zones dull green; reverse yellow at the margin to brown or purplish brown in center; conidiophores about 3.5 $\mu$  in diameter, with walls thin, apex swollen varying in length with zonal relationship, partly fasciculate; penicilli showing branches unequal 20 to 30 $\mu$  long metulae sterigmata and conidial chains at various levels; metulae about 11 to 14 $\mu$  long; sterigmata 10 to 12 by 2 to 2.5 $\mu$ ; conidia subglobose 3 to 3.5 less commonly 4 $\mu$ . Colonies grew very little at 30°C., or above but better at 20°C.

Another culture from the British Cotton Industry Research Association approaches the specifications of *P. stephaniae* closely. Certain observations on this strain may be noted. The zones were about 1 mm. in width; the outer zones were bright green with a bluish cast, quickly changing to green and passing into gray shades in center.

301. *P. palitans* Westling. Arkiv för Botanik 11: 53, 83-86, figs. 54 and 12. 1911.

Colonies in prune gelatine, green (C.d.C. 333, 313, 329, 309), in age dark gray green, not floccose, with narrow white granular scarcely wooly sterile margin consisting of creeping hyphae from which the conidiophores arise; reverse uncolored, or very pale yellow; gelatine slowly or only in part liquefied; hyphae rather coarse, 3 to 6.5 even to 8 $\mu$  in diameter; odor moldy; conidiophores arising from creeping hyphae, with range of 50 to 600 $\mu$ , but usually 90 to 300 in length by 4.4 to 6.5 $\mu$ ,

rarely up to  $8\mu$ , with walls smooth when young, often verrucose in old cultures, more often branched than in other species; penicillas 60 to  $175\mu$  long, figured as (1) branch or verticil of branches fairly appressed, (2) metulae, (3) sterigmata; metulae 12 to 16 by  $6.5\mu$ , with walls smooth or "uneven;" sterigmata about 9 to 11.5 by  $3.2$  to  $4\mu$ ; conidia at first pear-shaped, oval, or oblong (länglich), then becoming globose, to resume the broadly oval form when ripe, smooth  $4$  to  $4.7$  by  $3.6$  to  $4.3\mu$ , becoming  $7$  to  $8\mu$  in germination.

Species recorded by Westling as "common" and as having been received from Thom as a contamination of *P. decumbens*, hence also in America. He notes that it shows certain similarities to *P. solitum* and *P. conditaneum*, differs in the form and color of the conidia in which it resembles *P. viridicatum* whose spores are smaller and always globose (? C. T.).

Westling's designation of the change in shape of the conidia from initially oval to globose and back to oval or broadly elliptical must be doubted. It is probable that the mixture of broadly elliptical and subglobose conidia common in the genus mislead him at this point. Calcium oxalate crystals as columns or needles, often in bundles were recorded. Cultures grew poorly at  $30$  to  $31^{\circ}\text{C}.$ , changed litmus gelatine to a persistent red color, did not grow in 10 per cent tannin solution but grew in all other media tested with consistent green colors and absence of color production in the substratum.

The species is discussed in Biourge Monogr. La Cellule 33: fasc. 1, pp. 136-137; Col. Pl. XI, Cart. 355; Pl. XVIII, fig. 108, 1923. Biourge's no. 355 (our no. 4733.94) received in September, 1927, agrees in all essentials with this placing of *P. palitans* Westling; colonies grown upon slanted Czapek agar plates spreading, indistinctly zonate in the thin areas only, granular fasciculate at the margin with conidial areas bluish green at first then later deeper green, Russian green to stone green, (compare Ridgway XLI.); reverse colorless or pale yellowish; drops crystal, very large and abundant on deep areas of the plates; odor moldy, fragrant; conidiophores with walls rough or pitted, short to  $200$ - $300\mu$  or longer by  $3$  to  $4\mu$  in diameter; penicillus complexly branching  $60$  to  $80\mu$  long; conidia elliptical to globose, variably about  $4\mu$  in diameter.

*Sub-section 4. Glauca.* Colonies in gray green or glaucous and related shades rather than viridis or aeruginous. See Ridgway's tables XLI, XLVII, in which green is modified by addition of more gray than in Saccardo's viridis, and by the admixture of yellow and neutral gray not blue, in contrast to aeruginous. Such separation is largely arbitrary

but in the great number of cultures handled seems to bring together organisms that belong together.

This sub-section includes the great series of apple rot organisms, some of which are very destructive. In morphology they run from strains almost velvety with only vestiges of fasciculation in Czapek cultures to forms in which coremia are evident and sometimes predominante. These strains were probably included as the coremium producing varieties in Wehmer's (1893) and Sopp's earlier conceptions of *P. glaucum*. The sub-section name proposes the recognition of this for as nearly the *P. glaucum* group of organisms as we can approximate.

Literally hundreds of strains with individual differences when carried in parallel culture have been seen. These differences are so difficult to define that a stable nomenclature strain by strain seems impracticable at present. Identification upon quantitative studies of activity may sometime be possible.

Within the resources of a culture laboratory certain series may be established and some of the species already described assigned to them with some hope that identifications may be practical although the permanence of such division is problematical.

*Series: Crustaceum.* Fries described *P. crustaceum* in terms which have been widely interpreted as synonymous with *P. glaucum*. Both names have continued in use for indefinitely identified green Penicillia by various groups of workers to the present without established grounds in descriptive data, in exsiccati, or in fairly credible tradition to fix either name to a definite strain. Certain strains of the group constantly produce conidial masses which form plane or faintly ridged (zonate) surfaces composed of chains of conidia 100 to 500 $\mu$  in length packed together into masses and breaking off as angular sheets when the culture or natural substratum is jarred or tapped, thus giving the appearance of crusts which might justify the name. Such appearances are not confined to this subsection or even to this section since similar masses of conidia are found in *P. dierckxii* Biourge among monoverticillate forms, in *P. oxalicum* Currie and Thom among the velutinous Asymmetrica and in *P. viridicatum* series already discussed, but crusts are prominently produced in this subsection Glauca and it is therefore proposed to use Fries name "crustaceum" for a series of glaucous forms producing such crusts in contrast to the irregular and uneven masses produced by *P. expansum* on its fascicles or coremia or the masses that fall to powder in other sections.

As a type strain of this series we have taken a culture no. 5034.16

purified from a stock culture from Nobel's Explosions Company received from Scotland in January, 1929. Crust formation is beautifully represented by this organism. It shows fasciculation but only in rudimentary fashion for the most part, hence marks a favorable species to represent one extreme of this group (*Glaucy*) in which very great variation is constantly encountered. This organism is presented as a new species, *P. crustosum* Thom.

305. *P. crustosum* Thom, n. sp.

Colonies upon Czapek's solution agar in sage green and related shades, in age becoming brown shades such as cinnamon drab or Benzé brown Ridgway XLVI, azonate or indistinctly zonate in age appearing velvety but showing rudimentary fascicles at the growing margin, and occasional scattered fascicles longer, and becoming prominent in the center of older colonies, narrowly growing, about 200 to 300 $\mu$  deep in velvety appearing areas with the development of continuous crusts of conidial chains, which break off as irregular masses when struck or tapped; margin white 1 to 2 mm. in the young colony; reverse colorless or nearly so; drops colorless; most frequent in the older central area; conidiophores up to 300 or even 400 $\mu$  long, mostly much shorter, with walls rough pitted; penicillus consisting usually of the main axis and one branch variously up to 25 $\mu$  long appressed with few metulae 13 to 20 $\mu$  long and small groups of sterigmata 10 to 11 $\mu$  long by 2.5 to 3 $\mu$ , produced at more than one level in the penicillus; conidia about 4 $\mu$  in diameter or commonly slightly elliptical 4 by 3.5 $\mu$ , with a faint suggestion of pitting or roughening in the wall.

Type: No. 5034.16 purified from a mixture received from J. H. Birkinshaw, Nobel's Explosions Company, Ayrshire, Scotland.

*Series: Restrictum.* Colonies in gray green, to glaucous shades; zonate with zones narrow and fairly crowded.

307. *P. Blakesleei* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 441, 443, 444, Taf. 36. Zaleski's no. 253a.

Colonies upon neutralized Raulin with 10 per cent gelatine in petri dishes, quickly growing and becoming 6 cm. or more in diameter in twelve days, with gelatine at first softened then partly liquefied; thin, plane, velvety, definitely zonate throughout, with center raised like an umbilicus and loosely overgrown with secondary gray aerial mycelium;



margin (fimbria) white, loose, 1 to 2 mm. wide in the growing colony; in color at 6 days green, reverse plane, zonate; at margin C.d.C. 342 then to 338 and in center becoming 335 by the 10th day, passing 334, to dark fuliginous shades such as 140, 145, and 215 under various conditions in age; reverse zonate, in pale yellow greens, and yellows, to dirty orange browns; drops few small uncolored and developing mostly in the newer areas; liquefied gelatine almost uncolored; odor strong (intolerable); conidiophores straight or slightly flexuous, occasionally branched, 300 to 400 or even  $600\mu$  long, mostly 4 to  $5\mu$  in diameter, with all walls smooth; penicillus mostly 50 to  $60\mu$  long; branches straight or incurved 22 to 32 by 4 to  $4\mu$ , commonly 1 or 2 in number; metulae cylindrical or somewhat enlarged at base and apex, about 14 to 22 by 3 to  $4\mu$  in groups of 4 to 6; sterigmata 10 to 13 by 2.5 to  $3\mu$ , in verticils of 5 to 8; conidia smooth, globose or subglobose, at first about  $3.5\mu$  mostly 4 to 4.5 occasionally  $5\mu$ , in chains which break up in mounting.

Species isolated from humus under pine trees at a depth of about 5 cm. in the neighborhood of "Długa Gosłina" near Poznan in Poland. Zaleski classes it "Euconcentrica-classica." Our notes follow: Colonies upon Czapek's solution agar zonate, showing fasciculate conidiophores at the margin, forming a mass 300 to  $400\mu$  deep, with white marginal zone narrow (about 1 mm.), then a zone of bluish green, several zones in shades of bluish green to green, and a central area in gray green to gray tones with some overgrowth, (mouse gray, Ridgway LI); upon wort producing colonies in brighter green shades, more prominently zonate, and conspicuously radiately wrinkled, with center almost umbonate; reverse colorless then slowly showing slight tints of vinaceous (Ridgway XXVII, pale vinaceous pink); odor, indefinite or faint on Czapek, but strong, penetrating on wort; drops (leaving thin residues when dried up) central, very slightly yellowish; conidiophores 100 to 200 by  $4\mu$ , with walls delicately roughened or pitted; penicilli rather compact, with conidial chains more or less aggregated into columnar masses, consisting of branches at one or two levels, bearing metulae 12 to  $15\mu$  long, sterigmata up to 12 by  $3\mu$ , with conidia at different levels; conidia about 4 to  $4.5\mu$  in diameter.

Culture no. 5010.30 received as type from Baarn in July, 1928, contained two molds, one white or nearly so, and a green species which is described above and appears to comply with Zaleski's description well enough to establish a presumption as type although the identification can not exclude the possibility that the contaminated culture was actually in use in preparing Zaleski's description.

309. *P. porraceum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 188-189; Col. Pl. V, Cart. 403; Pl. IX, fig. 49. 1923.

Colonies on wort gelatine, forming a close felt with velvety appearing conidial areas, with very narrow white margin (0.2 mm.), bright green to olive; reverse in various shades of yellow and greenish yellow; liquefaction not reported on wort gelatine; conidiophore about 4 to 4.2 $\mu$  in diameter; penicillus 35 to 50 $\mu$  long, with all walls smooth, figured as main axis and one branch or a pair of branches each with a verticil of metulae, one or more of which occasionally bears a secondary verticil of metulae; branches 14 to 20 by 3 to 3.5 $\mu$  in twos or threes, sometimes reported; metulae 7 to 11 by 2 to 3 $\mu$ , in verticils of 2 to 4; sterigmata 8 by 2 $\mu$  abruptly sharp pointed, in verticils of 2 to 4; conidia at first ovate appendiculate, soon globose, 3 to 4 $\mu$ .

Biourge no. 403 (our no. 4733.98) was incompletely studied by him but regarded as belonging with *P. stoloniferum*; Biourge emphasizes narrow conidia bearing tubes in this group and the commonness of a so-called "connective" between conidia in the chain. This culture (4733.98) on Czapek's solution agar, produced colonies close felted floccose, with surface uneven up to 800 or even 1000 $\mu$  deep, dull green (celandine to artemisia Ridgway XLVII), becoming mouse gray in age; reverse and agar vinaceous, fawn color; odor doubtful; conidiophore about 4 $\mu$  in diameter, with walls delicately punctate or pitted (seen only with high magnification); conidia about 4 $\mu$ .

It was suggested as belonging with *P. stoloniferum* by Biourge, but description does not fit. Forms suggestive of this description have been contributed by pathologists studying decay of vegetables.

310. *P. aurantio-griseum* Dierckx. Soc. Scientifique Bruxelles 25: p. 88. 1901. See Biourge, Monogr. La Cellule 33: pp. 126-128; Col. Pl. I, Cart. 63; Pl. II, fig. 8; 1923.

Colonies on wort gelatine floccose, broadly zonate, blue green (C.d.C. 367), with white margin 2 mm. wide, coremia none; gelatine liquefied and the liquid orange red; reverse spotted with yellow to orange, to brown or orange red, odor none or very slight; drops uncolored or yellowish; conidiophore 4 $\mu$  in diameter (about 400 $\mu$  long on Czapek), with wall delicately pitted (high powers of the microscope), penicillus mostly 40 to 50 occasionally up to 85 $\mu$  long, figured as coarse and irregularly branching; branches 18 to 24 by 4 $\mu$  in twos or threes; metulae 10 to 12 or even 16 $\mu$  by 4, in threes; sterigmata 9.5 to 11 by 3 $\mu$ , in verticils of 2 to 4; conidia 3 to 4.5 by 2.5 to 3 $\mu$ , elliptical.

Biourge received this species (his no. 63, our no. 4733.7) from Kral in 1898 as *P. glaucum*.

Diagnosis based upon Biourge's culture no. 63 (our no. 4733.7). Colonies on Czapek's solution agar, floccose about 600 $\mu$  in depth, broadly zonate, gray green to bluish gray green; reverse and agar yellow to orange to brown; odor moldy, strong; conidiophore up to 400 by 4 to 5 $\mu$ , with walls delicately pitted (seen only when carefully examined with high magnification, suggested in one of Biourge's figures); penicillus with both primary and diverging branches followed by a secondary and unequal verticil or with a single compact verticil of branches then metulae and sterigmata; conidia 3 to 4.5 by 2.5 to 3 $\mu$ , elliptical.

This may well represent Sopp's *P. glaucum* in one of its many varieties.

*Series: Expansum.* Colonies in gray green or glaucous shades, fascicles typically in well marked zones and often showing definite coremia, especially when the substratum contains much sugar; coremia prominent on rotting apples, grapes, etc.

Conidia globose or subglobose.....Nos. 315 to 324.

Conidia elliptical.....Nos. 325 to 335.

315. *P. expansum* Link emended Thom; *Penicillium* rot of apples and allied fruits. See Link Obs., p. 17, 1809, and *ibid.*, *Coremium glaucum*, p. 19; *Icones* V, fig. 31; *Floccaria glauca* Greville, *Scottish Flora*, Pl. 301, figs. 1-4; *P. glaucum* Link (in part), *Species Plantarum* VI, p. 70, 1824; *Coremium vulgare* Corda (in part) *Prachtflora*, p. 54, Pl. XXV, figs. 3, 4, 17, 18, 19, 20, 21; Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 27, 28, fig. 1, 1910; *P. leucopus* Biourge *Monogr.*, p. 107, fig. 1, 1923. Our figures 60 and 61.

For original description see Chapter I. In this place Thom's (1910) description has been slightly emended to bring the terms used into harmony with the other species descriptions: Colonies upon gelatin and potato or bean agar, green becoming gray-green and slowly brown in several weeks (especially when exposed to light), with concentric zones tufted with short, loose, coremium-like aggregations of conidiophores, not over 1 to 2 mm. in height except in old cultures containing sugar, broadly spreading with broad white margin in growing colonies; reverse somewhat brown; conidiophores either very short lateral branches of aerial hyphae or very long (1 mm. or more), arising singly or grouped with others to form coremia; penicilli consist of 1 to 3 main branches bearing verticils of branchlets supporting crowded whorls of sterigmata

with conidial chains totalling 130 to 200 by 50 to 60 $\mu$  at base in cultures without sugar, with sugar continuing for some weeks to produce great numbers of conidia which come to form masses perhaps 1 mm. in thickness; sterigmata 8 to 10 by 2 to 3 $\mu$ ; conidia elliptical to globose 2 by 3.3 $\mu$  at first, later 3 to 3.4 $\mu$ , green, homogeneous, persisting in chains when mounted; colonies begin to liquefy gelatin slowly after about ten days and continue until it is completely liquefied. Grows readily and rapidly upon all common media.

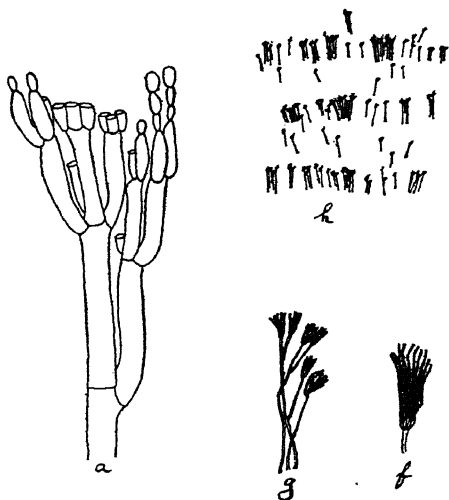


FIG. 60. *P. expansum* Link: a, detail of penicillus; f, sketch of penicillus; g, small group or fascicle of conidiophores; g, diagram of marginal zones of a magnified colony to show the zonate fasciculate groups of conidiophores with scattered simple ones (see fig. 9b for the massive type of coremium).

Occurs characteristically upon decaying apples and other pomaceous fruits, where old colonies often produce coremia 1 cm. or more in length and very large.

The author has preferred to continue the use of the name *P. expansum* instead of *P. leucopus* as proposed by Biourge for this series of strains. *P. expansum* was described by Link among the species assigned to his genus *Penicillium* at the time it was established hence offers a structural type (see Chapter I) for the genus which would be lost if the name must be abandoned since no one has the slightest idea what organism was ac-

tually in Link's hands when he described the other two species, *P. glaucum* and *P. candidum*.

When *P. expansum* was discussed in Thom's "Cultural Studies" in 1910, he did not appreciate the number of variations in habit, in coremium formation, in shades of color in the conidial area, in color in the reverse of the colony, and in odor production which would later be found. Literally hundreds of isolations belonging to this series have been made since then. Cultures have been received from correspondents in many lands and varying in number from 25 strains selected as separate and sent in one package to many single cultures sent in for identification or confirmation. Detailed study of the work of Wehmer, Sopp, Bainier, Biourge, Brooks with his coworkers, and many others, shows that the coremium-producing apple rot has been assigned to *P. glaucum* by many workers clear back to Link's *Genera Plantarum*, 1824. If that were all, it would be simple to accept *P. glaucum* for this species, but Brefeld's classic paper on *P. glaucum* with its sclerotia which slowly develop into ascus-producing perithecia introduces some other form.

Sopp remarks that *P. glaucum* is the species always found upon apples, but when he describes *P. glaucum* as he has it in culture he gives data which exclude the apple rot organism as we know it both upon apples and in culture. Bainier offers no figure or description for *P. glaucum* or for the apple rot species, but refers now and then to *P. glaucum* as a known form, while three strains nearly related to *P. expansum* were received in the Bainier collection, under names which certainly belonged to other forms. Wehmer (in Beitr. z. Kennt. Einheim. Pilze II, Taf. 1, fig. 5, 6, 7, 1895) figures the coremia of this species as *P. glaucum*, in connection with rotting fruit, but his uncertainty in the use of the name is clearly indicated by his description of one of these strains later as *P. variabile*, a culture of which was received directly from him. Uncertainties of usage thus surround the name *P. glaucum* clear back to Link's "Observationes" (1809), hence the name *P. expansum* as given in the same publication for the form upon rotting fruit is accepted in spite of the fact that Link had concluded by 1824 that the fruit rot was only one phase of a pleomorphic form, *P. glaucum*. *P. glaucum* is dropped for lack of any conception of what form Link had before him.

The name *Coremium glaucum* occurs fairly frequently in the literature for specimens upon rotting fruits which show coremia which we now know mostly if not all belong here.

Müller (in Fl. Danica Table 897 fig. 1) gives *Byssus scoparia* with

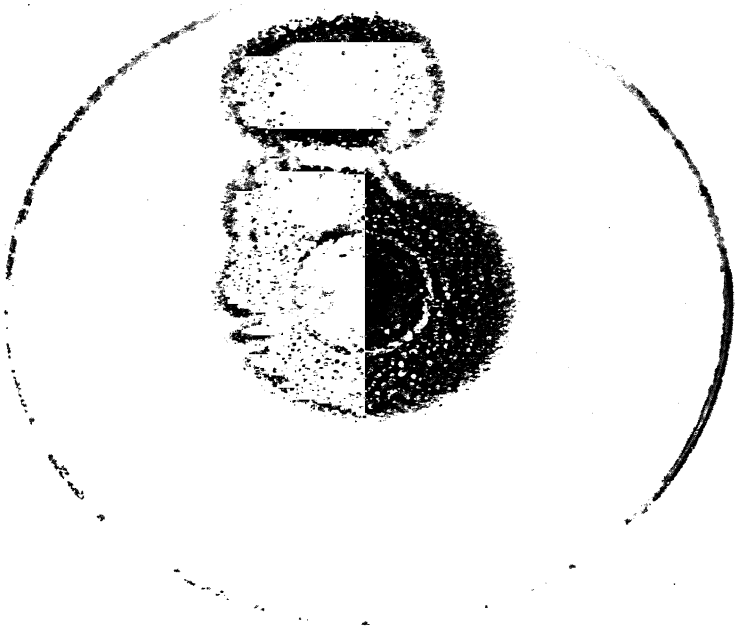


FIG. 61. *P. expansum*: Photograph of two large colonies with coremia forming interrupted zones.



figures which put the material definitely with the coremiform phase of *P. expansum*.

The specific names numbered from 316 to 324 and alphabetically arranged are used by various authors for organisms which appear to belong with the apple rot *Penicillium*. *P. elongatum* with its elliptical conidia may well be included. We have found it fruitless to attempt to attach one or the other of these names to particular forms, although the differences in description indicate a real range of variation among them. In a general way two subseries may be suggested because strains are often found which produce ridges of conidiophores and fascicles so separated by furrows or sterile zones as to be well described as sulcate, a condition probably typical of Biourge's euzonate species; in the same media other members of the series while showing well-marked zonation have no sterile furrow between the ridges. Even among the sulcate strains, the sulcate or euzonate condition is not always developed but appears only under favorable conditions of culture hence separation is not sharp and satisfactory. Tentatively then, the suggestion stands:

Colonies sulcate-zonate.....	<i>P. leucopus</i> subseries
Colonies zonate but not sulcate.....	<i>P. expansum</i> more strictly.

316. *Coremium albitopus* Secretan. Myc. Suisse III, p. 539-540. 1833.

Similar according to Secretan to *C. leucopus* Pers. Two varieties, *A* and *B* were described. In var. *A*, conidiophores were closely grouped forming a white stalked coremium with head at first white, then sea green, the whole appearing farinose to the naked eye, and being in length not more than twice the diameter of the head, with the penicillus divergent, 1/3 ligne in diameter. In var. *B* the head of the sea-green coremium was decidedly round, very large, borne on a thick compressed stalk, at first white, and then reddish and farinose, with the penicillus very short [Var. *B*. might have been *P. claviforme* Bainier—C. T.]. Variety *A* was found on starch paste and variety *B* on prunes; both are described in terms consistent with forms related to *P. expansum* as seen by us upon many different substrata but without details which would separate the strains studied by Secretan from each other or other members of the group. His observation emphasizes the different aspect of such a species when grown upon substrata presenting a sharp contrast in nutrient values.



317. *P. crustaceum* Fries. Sys. Myc. 3: 407. 1829. See *P. expansum* Link (no. 315) and *P. crustosum* Thom (no. 305).

This name perpetuates the idea that *Mucor crustaceus* of Linnaeus was one of these *Penicillia*. But even at that time Greville (1823) notes "*M. crustaceus* of Linnaeus, no mycologist would now venture to identify."

Fries' species was described in terms which would apply to any green *Penicillium*, but his form or variety  $\beta$  *coremium*, which he reported as seen upon rotting apples in autumn, and as identical with Persoon's *Coremium leucopus* (Myc. Eur. 1, p. 42) and Greville's *Floccaria glauca* (Crypt. Scot. t. 301) was clearly enough one of the forms we have discussed here and elsewhere as *P. expansum* Link. Subsequent references in the literature to *P. crustaceum* Fries are presumed by the users of the name to be this form, although many of the identifications are certainly questionable.

If the term *crustaceum* as applied by Fries was based upon the observation of masses of conidia which break off as crusts when the material is handled as appears to us now as probable, the name may apply to many forms since the appearance of these crusts is characteristic of some species on all media and other species upon rich media.

We have therefore introduced as representing a series "*crustaceum*", *P. crustosum* n. sp. no. 305. In this species the fasciculate character of the colony is noted only at the margin of the young and rapidly growing colony but the heavy "crusts" of conidia become prominent in the well developed colony.

318. *P. expansum* Thom. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 139-141; Col. Pl. II, Cart. 84; Pl. III, fig. 15. 1923.

Certainly not *P. expansum* Link in Thom, 1910.

Colonies on Raulin and wort gelatine, zonate, with coremia fairly abundant, green (C.d.C. 339, 343), surface granulate to the margin (with tufted or fasciculate conidiophores); reverse in various orange tones, irregularly distributed; gelatine liquefied but not discolored; odor nearly absent; conidiophore 3 to  $4\mu$  in diameter, with walls all smooth; penicillus about  $40\mu$  long, figured as either a verticil of branches bearing metulae or a single verticil of metulae, and in the figures symmetrical; branches 17 by 2.5 to  $4\mu$ , in twos or threes; metulae 9 to 12 by 2 to  $3.5\mu$ , in groups of 2 to 4; sterigmata 8.5 to 11 by  $3\mu$  in groups of 2 to 4; conidia elliptical, scarcely deciduous 3 to 4 by 2 to  $3\mu$ .

Biourge discussing his no. 84 (our no. 4733.55) finds it less common in Belgium than many others, but considers it a good species since he found it two or three times and compared it to Thom's culture (?) no. 14. He published the identification as correct, dropping the reference to Link and adding as a note that the absence of odor production created a doubt. Our transfer no. 4733.55 of Biourge's no. 84 did not suggest *P. expansum* to us, did not rot apples in moist chambers in which *P. leucopus* (4733.81), and our 4852 recently isolated were active rots. Disregarding our responsibility or lack of it for the culture held by Biourge as *P. expansum* Thom, *P. expansum* as understood by us is positively the coremium producing *Penicillium* which rots apples, hence either synonymous with *P. leucopus* or a closely related strain if separable. Biourge's description certainly does not describe the organism we received from him.

319. *P. glaucum* Link fide Wehmer. Beitr. z. Kennt. Einh., Pilze II, 1 p. 76-77; Taf. I, fig. 5 and probably fig. 6 and 7; Taf. II, fig. 16-22.

Species readily separated from related organisms by shades of color which are pure more or less dark leaf green (rarely blue green) in contrast to the gray-blue or bluish gray, or brownish green of other species, determination must be based upon young cultures, all parts except the conidia colorless; conidiophores 200 to 400 $\mu$  by 4 to 5 $\mu$ , with branching mostly prominently alternate; sterigmata 8 to 13 by 3 to 4 $\mu$ ; conidia purely globose, about 3 $\mu$  in diameter; sclerotia rare, 0.1 to 0.8 mm. in diameter.

Bainier (in Bull. Soc. Mycol. France 21: 126, 1905) refers to *P. glaucum* as a coremium producing form, to which he applies the descriptive term "gerbe" (sheaf, fascicle and bouquet) and reports conidiophores erect, more or less compacted together, fairly tall, then at a fixed level, diverging and forming their *Penicillium* type of fruits. This type is designated "bien connues."

Saccardo (Syll. XX, p. 277, 278, 279; 1911) gives one hundred and four citations of papers or series of papers dealing with *P. glaucum*.

320. *Penicillium kap-laboratorium* Sopp. La Cellule 36: 454. 1925.

Biourge mentions this species name as carried by his new no. 9 and synonymous with *P. leucopus* Persoon.

321. *P. leucopus* (Pers.) Biourge. Compt. Rend. Soc. Biol. Paris 82: 877-880, 1919; also Monogr. La Cellule 33: pp. 107-111, Col. Pl. I, Cart. 40, Pl. 1, fig. 1, 1923.

Synonym: *Coremium leucopus* Persoon, Myc. Europaea 1: 42, 1922; *P. expansum* Link of this book q.v.

Colonies on wort gelatine blue-green, zonate, with broad white growing margin, within producing gregarious, solid coremia with stalks white, round heads up to 1 to 2 mm. high, by 1 mm. in diameter arranged in the zones, coarser and larger coremia in age, or under special conditions, occasionally with heads sterile, with moldy odor strong; taste "intolerable"; drops uncolored, abundant; conidiophore  $3.5\mu$  in diameter, long, with walls smooth, penicillus commonly 50 to  $100\mu$ , occasionally up to  $160\mu$  long, figured as a main stalk with branches which often exceed it in length, producing sterigmata at various levels; branches 20 to 60 by  $3.5\mu$ ; metulae in twos or threes, 10 to 13 by  $3.5\mu$ ;



FIG. 62. *P. leucopus* (Pers.) Biourge: Diagrammatic radial section of margin of colony of Biourge's strain showing the sharply zonate arrangement of fascicles described by Biourge.

sterigmata in threes or fours 10 to 11 by  $3\mu$ ; conidia subglobose at first 2 to  $2.5\mu$  by  $2\mu$ , finally about  $3.6\mu$  by  $3.4\mu$ .

Biourge's culture no. 49 (our no. 4733.81) differs little from the usual types of apple rot *Penicillium* found in America (see fig. 62), but in several lots of cultures of American origin, carried together upon Czapek's solution agar in 1929, *P. leucopus* (4733.81), 5050.45, 4975.162, 5034.50 from Scotland and others produced colonies in which zones or ridges of conidial masses were quite sharply separated by furrows of sterile or nearly sterile mycelium in contrast to the more usual form of the *P. expansum* type of colony in which zones are marked by ridges of taller conidiophores and fascicles, separated by shallower zones but all well covered with green conidial masses. Even this distinction is often obliterated by changed conditions in subsequent cultures. It remains, therefore, problematical, whether we have a real basis of separation of the various strains. All such cultures produce deep but irregular and uneven masses of conidia which break off as such irregular masses not in crusts as seen in the "crustaceum" series already discussed.

While these strains differ in minor cultural reactions, no differences

(omitting the euzonate feature) deemed adequate for describing them as separate species appear in our records. In examining hundreds of isolations with this general morphology a considerable range of variation is found in the shade of bluish green to green in the conidial areas, in the amounts of color developed in the substratum which varies from reverse and substratum colorless or nearly so to strains with more or less yellow, or with yellow to brownish, or with reverse passing quickly to brown or even reddish brown. Even greater variation appears in the abundance and definiteness of coremium production in laboratory media. When such cultures are kept continuously in the laboratory with repeated transfers, many, if not all of them, tend to reduce coremium production to vestigial tufting or sporadic production of definite coremia under difficultly determinable conditions. The two extremes showing marked contrast as originally isolated tend toward a common aspect after many transfers. While some of these differences may justify using a name if found to correspond with a published description, we have not felt justified in describing species among them because of the general consonance in structure and habit. Biourge gives the synonymy of *P. leucopus* as including *Floccaria glauca* Greville, *P. juglandis* Weidemann, *P. elongatum* Dierckx, *P. variabile* Wehmer, *Coremium vulgare* Corda in Prachtflora tab. XXV, figs. 1, 2 and 3, but not the others and possibly *P. glaucum* var. *pallidum* of Sopp. We have already questioned the identity of *P. elongatum*. Our own cultures from Wehmer's *P. variabile* agree with this placing. Weidemann's organism is excluded by his description and has been placed elsewhere. Greville's *Floccaria* and Corda's *Coremium vulgare* may readily have included members of the group but no one can safely identify either of them as particular strains.

Persoon's *Coremium leucopus* remains—this also may easily have been some member of the series of apple rot forms but it is as equally unsafe to hazard a guess as to whether he had one of this group or *P. claviforme* of Bainier.

*P. malivorum* Cifferi. Riv. Patol. Veget. 14: 77. 92. 1924.

Free translation and rearrangement (C. T.) of Latin diagnosis: Colonies on wort gelatin with granular powdery appearance, at first greenish glaucous, then fuscous green or aeruginous to olivaceous green, producing abundant coremia with stalks white or gray, 1.5 to 2 by 2 to 3 mm. (given as  $\mu$  by Ciferri but presumably a typographical error); reverse at first cream, then ochroleucus, ochraceus, to ferruginous; gelatine strongly liquefied; odor characteristic; conidiophore 7 to  $7.5\mu$  in diameter, with all cell walls smooth; penicillus up to 33 to  $35\mu$ , or includ-

ing the branches 45 to 60 $\mu$  long; branches in 2's or 3's, 10 to 14 $\mu$  long; metulae 6 to 9 by 2.5 to 3.5 $\mu$ , 4 to 6 commonly 5 in the verticil, with apex clavate or somewhat vesicle-like; sterigmata subcylindrical to subclavate, 17 to 21 by 3 to 4 $\mu$ , usually 19 by 3.5 $\mu$ ; conidia globose 2.5 to 4 $\mu$ , usually about 3.5 $\mu$  diam., greenish hyaline, persistent in long chains.

Habitat: Decaying quinces (*Cydonia vulgaris*) in Italy.

Ciferri evidently had a member of the *P. expansum* group, showing coremia abundantly, coarse smooth stalks, and reverse ferruginous.

323. *P. plumiferum* Demelius. Verhandl. Zool.-Bot. Gesellsch. Wien 72: 76, fig. 5, (1922) 1923.

Colonies in prune gelatine velvety or floccose, blue green (C.d.C. 366, 367, 347, 348); conidiophores with walls smooth, arising from creeping or submerged hyphae, up to 2 mm. by 3 to 4.5 $\mu$ , often (partly) aggregated into plumose coremia spreading from the middle upwards to form an umbrella-like head; penicillus figured as central axis and opposite branches bearing metulae and sterigmata; metulae 9.6 to 13 $\mu$  by 3 to 3.6 $\mu$ , usually in threes; sterigmata 7.2 to 8.4 by 2.4 to 3 $\mu$ , usually in threes or fives; conidia ellipsoid 2.6 to 3 (3.8) by 2.4 to 2.5 $\mu$ .

Species found in dried leaves of *Beta vulgaris* var. *ciclae*, at Schongeebern 1917, and regarded by Demelius as near *P. expansum*, from which it was separated on account of smaller conidia, greener color, the abundance of its coremia, and absence of characteristic odor.

It is not difficult to believe that some member of the *P. expansum* series may satisfy this description as seen in the following cultural notes: No. 4933.1 from F. D. Heald, at Pullman, Washington, in certain cultures gave the measurements and reactions of this species, with colony color on Czapek's solution agar near *Gnaphalium* green, Ridgway XLVII, and giving the typical odor of *P. expansum* only after sixteen days of growth. Retransfer of this culture parallel with No. 4733.81 the type of Biourge's *P. leucopus* gave identical colony characters while a transfer of 4933.2 which in its first form appeared to be *P. expansum* gave the few distinguishing colony characters of *P. plumiferum*. We do not, therefore, feel justified in placing confidence in these minor cultural differences as characters of species.

324. *P. variabile* Wehmer, Mycol. Centralb. 2. p. 195-203. 1913.

Compare Ber. deut. bot. Gesell. 31: 210-235, 1913; Meyer, R. Apotek. Ztg. 38: 763, 1913; also, Mycol. Centralb. 4: 72-76, 1914.

In Wehmer's description: Colonies on wort gelatine at first bluish green then gray green even to chrome-green (uniformly colored on all

media), becoming gray brown to brown in age on wort gelatine, aerial growth characterized by the development of coremia with colorless stalks, loose or compact, from small, scarcely definite, to 1 cm. in height, commonly with abundant simple conidiophores filling the interspaces; reverse from colorless to irregularly or commonly unevenly reddish yellow to deep orange; gelatine liquefied; sugar media soured; odor reported as none or slightly musty; conidiophores with walls smooth, simple or branching from the coremia; penicillus 30 to  $50\mu$  in length; metulae 10 by  $3.3\mu$ ; sterigmata 8 by  $2\mu$ ; conidia 2.4 to 3.2 averaging about  $2.8\mu$  in diameter; perithecia not found.

Species observed as a cause of rot in apples and oranges, not attacking raw potatoes, lemons or onions.

Culture no. 3551 received directly from Wehmer was a strain or race so closely related to *P. expansum* as to make separation difficult. Biourge, Monogr., p. 107, reports this species as certainly *P. leucopus*.

Cultures received under the name from certain other German sources in 1924 to 1926 have been races of *P. chrysogenum* hence the substitution of this organism for Wehmer's species has become complete in certain laboratories. Our own stock culture reexamined in 1929, showed the same replacement; this raises the doubt as to its original purity—i.e. Biourge evidently received the same culture we did, recognized the dominant organism in it as the coremiform or fasciculate species called by him *P. leucopus* and passed it to "stock" without critical purification, just as we did.

"*P. varians* Munk-Wehmer 1913" appears in Biourge's "List onomastique." Munk in his papers (Mycol. Centralb. 1 (12): 387-403, 1912), discusses a *Penicillium* without naming it. Wehmer in describing *P. variable* definitely identifies it with the coremium-producing species discussed by Munk. Evidently *P. varians* is a misprint. "*P. variable* Westling" also appears in Biourge's list but we can not find that Westling used the name.

*Conidia elliptical.*

325. *P. elongatum* Dierckx. Soc. Scientifiques Bruxelles 25: 87, 1901.

Synonym: (?) *P. leucopus* (Pers.), Biourge fide Biourge.

Colonies quickly growing, green then brown, characterized by the aggregation of conidiophores into coremia; reverse orange changing to brown and coloring the substratum brown; drops of exuded liquid abundant; penicillus up to  $60\mu$  long; sterigmata 3 to 4 in the verticil; conidia oval 3.5 by  $2\mu$ , separating readily.

Biourge with all Dierckx's data before him reported this to belong with his *P. leucopus* ignoring the ellipticity of the conidia reported by Dierckx. While ordinarily inclined to throw nearly related strains together, we have occasionally encountered a strain of the *P. expansum* series with conidia distinctly elliptical. Dierckx's name may perhaps be let stand to mark this as an extreme of variability in this series with its multitude of diverging strains. Our notes upon one of these strains follow (no. 5001.25) isolated from a cucumber, Evanston, Illinois, contributed by Prof. A. W. Povah; colonies upon Czapek's solution agar, near sage green (Ridgway XLVII), fairly broadly spreading, forming felts and areas of conidia usually less than  $500\mu$  deep, thinner at the margin, outer 1 to 2 mm., white succeeded by a loosely fasciculate light green (falsely bluish) zone, passing to green tones, with central area unevenly overgrown with filamentous masses obscuring fasciculation and zonation at first but later becoming ridged zonate; reverse colorless or pale lemon yellow in partial zones; odor indefinite; drops small variously distributed; conidiophores 3 to  $3.5\mu$  in diameter, separate or in fascicles; penicilli with compact branching system and cells full of vacuoles, consisting of main axis appressed 20 to  $40\mu$  long, secondary branches up to 18 to  $20\mu$ ; metulae up to  $15\mu$  long; conidia about 4 to 4.5 by 2.5 to  $3\mu$ , elliptical to subfusiform.

Differs from *P. expansum* primarily in the ellipticity of the conidia and lack of odor. Similar forms from widely separate sources are not rare.

Some other cultures with the elliptical conidia and lacking the characteristic odor of *P. expansum* have the same slight differences in shade of green and in color or intensity of color in reverse. Such observations support the maintenance of the name.

*Series: Italicum*—nos. 326, 327, 328.

326. *P. italicum* Wehmer. In Hedwigia 33: 211-214, 1894; see also Beitr. z. Kennt. Einh., Pilze II, 1, pp. 68-72; Taf. I, fig. 1-3, Taf. II, fig. 1-10, Jena, 1895. Compare fig. 63.

Colonies on infected citrus fruits with mycelium superficial or creeping, bluish green to gray green, during the growing period showing a white sterile marginal zone, in which fascicles of conidiophores are more or less evident, varying with the variety or strain; conidiophores on sugar solutions, up to  $250\mu$  long by about  $4\mu$ , colorless, erect; penicillus about  $50\mu$  long, consisting of main axis and three or four branches alternately or variously produced at the upper nodes of the stalk; sterigmata about

10 by  $3\mu$ ; conidia elliptical, 4 to 5 by 2 to  $3\mu$ , at first definitely cylindrical and only slowly becoming elliptical; sclerotia globose up to  $300\mu$  in diameter, smooth, brown, fairly uniform in appearance, consisting of polyhedral thick walled cells white except for the outer layer, without further development; commonly their structures are surrounded by a gray fibrous felt of sterile hyphae.

Species rarely found except on citrus fruits where it produces soft rot. It grows well on many media. Literally hundreds of observations of this species in its effects upon oranges have been made. Many of them

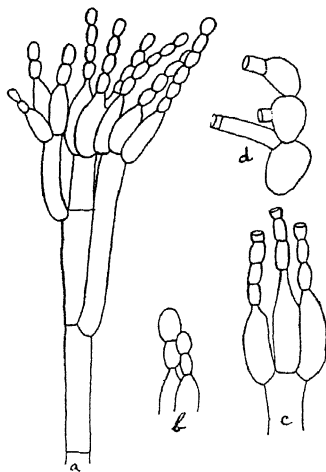


FIG. 63. *P. italicum* Wehmer: Detail figure and germinating conidia; c, the characteristic almost oidium like formation of the conidia

have been verified by microscopic examination. Our conception of Wehmer's species was based upon rotting oranges collected in the market in Hanover and carried to Wehmer's laboratory. Subsequent studies in America fully justify his discussion in 1895.

Wehmer described sclerotia in *P. italicum* but failed to find ascospores. Thom (1910) also reported sclerotia but failed to describe them adequately. In the subsequent examination of many lots of rotting oranges, a morphology harmonizing with Wehmer's description was seen regularly and a culture occasionally isolated but no sclerotia appeared in the cultures studied. Schwartz (1926) found a strain of *P. italicum* (identification confirmed by Wehmer) which produced clumps of



sclerotia after three weeks' growth upon oranges. The sclerotia develop most abundantly under conditions in which conidium formation is suppressed or greatly reduced. The mass of sclerotia formed in his cultures was separated, washed, and preserved on filter paper at different temperatures and percentages of humidity. After twelve weeks at room temperature and humid conditions a few sclerotia had become brittle, and when crushed, showed loose hyphal networks and a few free ascospores, some unbroken asci and ascogenous filaments. These ascospores are smooth walled, measure in profile  $2.6$  by  $3.9\mu$ , this includes the width of the "ring;" viewed on the flat side they are nearly circular, about  $3.8\mu$ , of which the ring or band constitutes about  $0.5\mu$ . Single ascospores cultivated in wort agar tubes reproduced the original green *Penicillium* which again produced sclerotia. The author concludes that *P. italicum* is homothallic. Although we have tried many times, we have obtained no ascospores from our transfer of Schwartz's strain.

327. *P. aeruginosum* Dierckx. Soc. Scientifique Bruxelles 25: 87. 1901. Regarded by Dierckx as possibly *P. olivaceum* Wehmer at that time but later in unpublished notes found identical with *P. italicum* Wehmer. In Biourge Monogr. La Cellule 33: pp. 121-123; Col. Pl. I, Cart. 123; Pl. I, fig. 6. 1923.

Biourge gives as a possible synonym *P. divergens* Bainier and Sartory; we do not agree but on the basis of the description regard that species as correctly indicated by its authors to be related to *P. granulatum*.

Biourge gives: Colonies on wort gelatin in the zonate group, floccose or lanose, more or less wrinkled, bluish green, with coremia developing at the margin; reverse yellow to orange shades; conidiophore smooth,  $3.2$  to  $3.5\mu$  in diameter; penicillus  $40$  to  $60\mu$  long, figured as main stalk and branch or branches fairly closely appressed, and producing metulae and sterigmata at more than one level; branches  $16$  to  $20$  by  $2.5$  to  $3\mu$  in pairs; metulae  $10$  to  $12$  by  $2$  to  $3.2\mu$ , in verticils of  $2$  to  $6$ ; sterigmata  $10$  to  $11$  by  $3$  to  $4\mu$ , in verticils of  $2$  to  $6$ ; conidia globose or ovate,  $3$  to  $4.5$  by  $2.5$  to  $3.2\mu$ .

Biourge lost his culture before publishing his monograph. He reestablished *P. aeruginosum* as a coremium producing species after careful discussion of the failure of Wehmer, Thom and Weideman to find coremia in their cultures. Their descriptions were obviously based upon too few examinations and too few isolations from original material. We have isolated many strains since 1910, and found many of them to produce coremia especially at the margin of old cultures. There are clearly

a considerable number of strains of blue green *Penicillia* which produce soft rot of oranges but which show measurable differences in cultural characters—such as shades of color in conidial areas, development, location and abundance of coremia, colors, in reverse and in substrata, probably also in their destructive effects upon citrus fruits but thus far no comparative study of strains has been published. Without such a comparative study we are unable to separate *P. aeruginosum* from *P. italicum*, if there is ground for such separation. We are inclined to believe that Dierckx was correct in identifying this species with *P. italicum* Wehmer and to find no justification either in Biourge's notes or his diagnosis for reviving *P. aeruginosum*.

328. *P. ventuosum* Westling. Arkiv för Botanik 11, pp. 57, 112–114, figs. 26, 67. 1911.

Colonies in prune gelatine, lanose, gray green (C.d.C. 347, 322, 372) after a month or more brown, often partly overgrown with white mycelium, with a broad wooly white margin in which are small prostrate coremium-like columns of hyphae variously woven together, suggested as resembling *P. africanum* probably in these ropes and *P. italicum* by their marginal position; reverse uncolored to gray at first, later clear brown; gelatine quickly liquefied, with alkaline reaction, with columnar calcium oxalate crystals abundant in the medium; odor scarcely detectable; conidiophores arising from submerged hyphae or as shorter branches of aerial hyphae, erect, smooth, 75 to 700 by 3.2 to 4.8 $\mu$ , figured as forming a single verticil of clavate metulae, but described as occasionally vesicle-like at the apex and producing sterigmata only; metulae 12 to 16 by 3.2 to 4.8 $\mu$ ; sterigmata 7 to 9.6 by 2.4 to 3 $\mu$ ; conidia smooth or slightly verrucose, elliptical then oval to subglobose 2.8 to 3 by 2.2 to 2.7 $\mu$ , swelling in germination 4 to 5 $\mu$ , penicillus 45 to 120 $\mu$  long.

Species found but once on roots of *Valeriana officinalis* L. It grew poorly at 30 to 31°C. Cultures grew well in malt-extract-gelatine, plum gelatine, potato, maranta starch, citric acid solution, and bread. In milk no conidia were produced but white to reddish mycelium contrasted sharply with the colorless fluid.

A culture (our no. 2547) received from Westling as *P. ventuosum* was studied but found unsatisfactory in comparison with his own description (p. 113) in which he found in the margin of his older but still growing colonies, prostrate coremia which in our experience characterize certain strains of the *P. italicum* series but are not found in other groups. These structures apparently puzzled Westling himself and lead him to

question whether his organism belonged with *P. africanum*, a funiculose species which he evidently had in culture, or *P. italicum*, one strain of which he evidently had cultivated without encountering these structures. Recent work (unpublished) by McCulloch and Thom in connection with *P. gladioli* lead us to recognize much greater variation among the *Penicillia* causing blue green rot of citrus fruits than Thom or Wehmer indicated in their publications. Study of the series remains incomplete but we are convinced that Westling had a strain of *P. italicum* perhaps an impure culture when he described *P. ventruosum*. Biourge in his Monograph, p. 190, says he has not seen the species but gives a Latin diagnosis in his own terminology from data given by Westling.

*Colonies in gray green shades; conidia elliptical; not on citrus fruits and diverging from the P. expansum series in habit.*

*P. juglandis* Weidemann. Centralb. f. Bakt. etc., 2 Abt. 19: 683-687; fig. 2. 1907.

Colonies on glucose gelatine or wort-gelatine, gray green to dark green, at first producing a surface growth of simple conidiophores but with a "granular" appearance (beginning of coremia), later in these conidial areas producing coremia up to 1 to 2 mm. in diameter and 1 to 2 mm. in height, at first white, then green; reverse white to yellow or reddish yellow; glycerine solution and sugar solutions colored yellow; vegetative hyphae 2.5 to 3 $\mu$ , abundant clear or crystal drops appear in nearly all cultures; conidiophore 3 to 3.5 $\mu$  in diameter, little branched with apex of the stalk and every branch enlarged, bearing metulae or sterigmata; metulae often in threes, sterigmata mostly 3 or 4 in the verticil, up to 12 $\mu$  by 2.5 to 3 $\mu$ , somewhat rounded toward the base and abruptly narrowed to a rather broad conidia-bearing tube; conidia elliptical, at first 2.7 by 1.9, when ripe 2.7 by 2.3, wall single but a connective or bridge is commonly seen between ripe conidia in the chain.

Species found in a walnut in a fruit store at Keil, Germany. Extensive cultural experiments recorded include tolerance of 25 per cent tannin in the culture solution, the production of oxalic acid and persistence of an acid reaction for a long period in cultures, the production of brown mycelium but no green color upon milk with the complete digestion and reddening of the milk itself.

Biourge placed the species among the synonyms of *P. leucopus*. Since Weidemann's culture has not been seen we are left to our judgment in reading descriptions and we have included it here as a possible form

with more pronounced coremium formation than the usual forms of *P. expansum*.

*P. schneeggii* Boas, Myc. Centralb. 5: 73-83. 1914. See also Centralb. f. Bakt., etc., 2 Abt. 44: 695-696. 1916.

Colonies upon all solid media and at the edges of liquid media, characterized by the formation of coremia 2 to 12 mm. in length which are at first often short and radiating from central points, later feathery and often sterile, conidial masses green; reverse yellow at first on sugar media (associated with acidity) becoming reddish to red in age (with developing alkalinity); sterile hyphae 3 to 4 $\mu$  in diameter; odor of some ester produced; conidiophores mostly branching from the coremia about 4 to 5 $\mu$  in diameter with walls granulate or punctate, described as producing 2 equal branches 14 to 25 $\mu$  in length, at the apex (? main stalk and 1 branch probably C.T.) then 2 verticils of branches or metulae, primary 13 to 21 $\mu$  in length and secondary 7 to 13 in length and bearing sterigmata 8 to 12 $\mu$  in length; conidia at first elliptical then toward globose 2.5 to 2.8 $\mu$  in long axis, swelling in germination to 5.5 or 6 $\mu$ , mostly developing one germ tube.

Species found upon a chestnut associated with Botrytis at Weihestephana, Germany. Cultures grew well upon most laboratory media, with a temperature range of 3 to 32°C. but only slow growth at 32° and no coremia above 31°C.

Boas placed his species close to *P. corymbiferum* Westling on account of its granular walls accompanying coremium production and color production, as separate from *P. granulatum*, *P. claviforme* and *P. duclauxii*; *P. duclauxii* has smooth conidiophore walls and conidia delicately granular.

Bourge in 1925, reported a culture as received from Pribram to be a mixture of *P. claviforme* and some Scopulariopsis. He gave it in his "list onomastique" as *P. (corem.) schneeggii*.

In January, 1929, we received a culture labeled *P. schneeggii* from Nobel's Explosions Company through Dr. J. H. Birkinshaw. Upon Czapek's solution agar this produced colonies zonate 400 to 500 $\mu$  deep, plane or umbonate toward the center with margin spraying out in lines as radiating submerged hyphae which with the branching system produced lines of conidial fruiting and gave a markedly stellate appearance to young colonies; (in centers occasionally developing branching coremiform or Isaria-like structures often perfectly white, again showing some conidial covering; at times several millimeters high—see note

below); conidial areas olive gray; reverse yellow, orange, orange red, or blackish brown in age; odor strong, suggesting *P. digitatum*, persistent for at least one month; conidiophores produced commonly in clusters suggesting fascicles, few over  $100\mu$  long and about  $4\mu$  in diameter, with walls granular or almost spinulose; penicilli with few branches and producing tangled chains of conidia, suggesting *P. digitatum* in appearance; sterigmata 10 to  $12\mu$  long, few in the verticil tapering gradually; conidia about 4 by  $3\mu$ , ranging from a little smaller to a little larger.

When the feathery white coremia described by Boas began to appear the identity of Boas' type was no longer doubted, but its purity began to be questioned. This was followed up by careful transfer of bits of the white feathery growth which grew into pure white to cream colored *Irpe*x-like colonies, with densely crowded erect coremium like stalks, entirely sterile so far as studied. Similar transfers designed to remove conidia only tend to produce colonies free from these "coremia." We are compelled to believe that Boas' type culture was contaminated but that free from the coremiform contaminant it is still a good species and belongs about at this place in a scheme of classification. This species description emended by leaving out the bracketed clauses, describes our cultures as they appear at the present stage of purification.

*Series: Urticae-patulum nos. 331 to 335*

331. *P. urticae* Bainier. Bul. Soc. Mycol. France 23: 15-16; Pl. IV, figs. 1-5. 1907.

Synonym: *P. flexuosum* Dale, in Biourge and in Ann. Mycol. 24: 137. 1926.

Colonies on licorice sticks, at first light green, in age dark gray green; conidiophores partly separate and several branched above, partly fasciculate or coremiform and when in fascicles showing few branches irregular in length and arrangement; penicillus varying from comparatively simply branching with sterigmata 1 to few in the verticil to complex superposed verticils and masses of conidia; sterigmata rather short with length 2 to 4 times the diameter; conidia about  $2.8\mu$  in diameter, swelling in germination to 8 to  $9\mu$ , becoming several vacuolate and emitting one or two moderately sinuate hyphae which are also vacuolate.

Culture no. 4640.455 received as *P. urticae* in the Bainier collection seems to satisfy Bainier's figures and description sufficiently well to be accepted as a type culture. An identical culture from Miss Dale (our

2694) discussed as *C*<sub>1</sub> and *C*<sub>13</sub> in Ann. Mycol. 12: p. 44., 1914) has recently (Ann. Mycol. 24: 137. 1926) been named by her *P. flexuosum*. Two other cultures no. 4359.105 and 113 were received from Brierley at Rothamsted. One culture of American origin was received from Kopeloff in Louisiana (our no. 4282.23) in 1918; another no. 4894.7 labeled *P. cyclopium* as received from Gilman and Abbott proved to be *P. urticae*.

The following characterization is based upon our own observations which began with Miss Dale's culture (our no. 2694) in July 1912: Colonies on Czapek's solution agar with margin floccose, white, then slowly gray green (Ridgway XLVII, ggy, Gnaphalium green), spreading, usually zonate, with a granular or mealy appearance due to arrangement of conidiophores in crowded fascicles or coremia; reverse slowly cream to somewhat flesh color or reddish brown, sometimes with slight discoloration of the agar beyond the margin of the colony; drops abundant colorless; odor peculiar, characteristic of this species only; conidiophores 3 to 3.5 $\mu$  in diameter, more or less flexuous, separate or aggregated into fascicles (coremia) up to 1 mm. long; penicillus including a terminal verticil of metulae and more or less diverging branches from 1 to 3 of the succeeding nodes; sterigmata mostly short about 5 $\mu$  (to 7 $\mu$ ) by 2 $\mu$ , and frequently deciduous in age; conidia more or less elliptical about 2.8 to 3.2 (occasionally 3.5 $\mu$ ) by 2 to 2.5, rarely to 3 $\mu$  to subglobose, smooth, almost colorless by transmitted light (Zeiss apochromatic), swelling in germination to 5 $\mu$  and showing a large vacuole, while emitting a single germ tube; colonies liquefy gelatine rapidly. Upon apples produced a small area of rot (organism recovered by transfer), upon orange a necrotic area in the peel only.

332. *P. flexuosum* Dale, incorrectly attributed to Westling on p. 103 and in Col. Pl. XI, Cart. 359 of Biourge, published in Biourge Monogr. La Cellule 33: fasc. 1, pp. 264-265; Col. Pl. XI, Cart. 359; Plate XIX, fig. 110; 1923.

See also *P. urticae* Bainier.

Not *P. flexuosum* Preuss 1851.

Colonies on wort gelatine, restricted in growth, at first pale bluish gray green, then yellowish green, through a reddish or pinkish color especially at the margin at length to fuscus in age; coremia none; reverse pale ochraceous or orange; odor ethereal peculiar; conidiophore 2 to 2.5 $\mu$  figured as flexuous but not described; penicillus about 15 to 20 $\mu$  or double length when branched, with all walls smooth; branches occa-

sional, irregular in measurements; metulae 6 to 13 by 0.5 to 2 $\mu$  (?C.T.), commonly small below, enlarging upward, single or in twos, threes, or fours; sterigmata 7 to 10 by 2.5 $\mu$ , in groups of 2 to 6; conidia oblong to globose 3 to 4.5 by 2.8 to 4 $\mu$ .

Biourge's no. 359 (our no. 4733.62) appears to be no. 14 (C13) which was partially described in Miss Dale's Fungi of the Soil II, Ann. Mycol. 12: 44, 1914. This culture was our no. 2694, and is identical with no. 4640.455 from the Bainier collection described as *P. urticae* Bainier.

The name *P. flexuosum* is not in Westling and not in Dale's 1912 and 1914 papers but is proposed by Dale in Ann. Mycol. 24: no. 1/2, p. 137, 1926. In any case, the name is untenable because previously used by Preuss in 1851 (see no. 651).

*P. aeruginosum* Demelius. Verhandl. Zool.-Bot. Gesellsch. Wien 72: 76-77, fig. 6, (1922) 1923.

Not *P. aeruginosum* Dierckx. Probable syn. of *P. urticae* Bainier.

Colonies on potato plugs, velvety lanose, gray green near D.d.C. 297; conidiophores 2 to 3 mm. by 3 to 3.6 $\mu$ , aggregated into coremia, with heads (C.d.C. 367, 368, 293, 318) bluish green then gray green, penicillus about 112 by 30 $\mu$ , compact (geschlossen); metulae clavate 7.2 to 14.4 by 2.4 to 3.6 $\mu$ , usually in twos; sterigmata fusoid 7.2 to 10 by 2 to 2.4 $\mu$ , mostly in twos or threes; conidia ellipsoid to subglobose 3 to 3.6 by 2.4 to 3 $\mu$ .

Species found upon oil-residue, Vindobon, 1918. Without having seen Demelius' culture this species belongs by description very close to *P. urticae* Bainier.

334. *P. patulum* Bainier. Bull. Soc. Mycol. France 22 (fasc. 3): 208, Pl. XI, figs. 14-17; also *ibid* 23; Pl. V, figs. 10-16, 1907.

Bainier named and discussed his species as found upon excrement of sheep, using terms which others failed to use successfully in identifying other strains as found. The characters given were: Colonies showing a blue greenish shade different from previously known species; conidiophores fairly uniform in diameter from base to apex but differing in different individuals, figured and described as flexuous, i.e., curved or undulated; and commonly anastomosing at points of contact producing short connecting filaments; penicilli irregularly and divergently branched, with terminal "umbels" (groups of metulae, sterigmata and conidial chains), conidia recorded as very variable, oval when young, round when ripe and about 2.8 $\mu$ —figured in chains as elliptical, however.

Culture no. 4640.451 from the Bainier collection labeled *P. patulum* proved to be *P. paxilli* (synonym, *P. stoloniferum* Thom); culture no. 4640.454 labeled *P. puberulum* corresponded well enough with Bainier's *P. patulum* to establish the presumption of identity and even the probability of type material. We have since obtained apparently identical organisms from other sources. Our characterization based upon this strain follows: Colonies upon Czapek's solution agar in various shades of gray, dawn gray, or close to gnaphalium green (Ridgway XLVII), restricted or slowly spreading in the substratum, white margin rather broad, often 3 mm., becoming tardily and narrowly zonate in later stages of growth, fasciculate with fascicles crowded; odor indefinite or wanting; reverse yellow to salmon or rosy salmon; conidiophores partly in fascicles partly simple, walls smooth, undulate or sinuate, branching toward the apex and the branches diverging (whence the name: *patulus*—open) and bearing almost independent penicilli; individual penicilli either consisting of verticils of metulae or one or more branchlets then verticils of metulae 15 to 20 $\mu$  long, closely packed; sterigmata 12 to 13 by 2 to 3 $\mu$ , closely packed, falling off in mounts in old cultures; conidia almost hyaline, smooth, up to 3 to 3.5 or 4 $\mu$  by 2.5 to 3 with some ellipticity usually in the ripe spores, figured by Bainier as much enlarged in germination and putting out one or two tubes which show abundant vacuoles.

Colonies grown in gelatine produce abundant liquefaction with orange to reddish coloration.

Species closely resembles *P. urticae* Bainier but lacks the characteristic odor of that species, and produces somewhat less abundant growth. While the two species are fairly closely related, other strains with similar differences have been encountered.

335. *P. polonicum* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et Nat. Ser. B, 1927, pp. 445, 446, 447, Taf. 38, Zaleski no. 1431b.

Colonies on neutral Raulin with 10 per cent gelatin in petri dishes, thin, plane, quickly growing, becoming more than 5 cm. in diameter in twelve days, with the gelatine becoming strongly liquefied about the tenth day; appearance velvety, entirely plane, regularly concentrically zonate, overgrown in the raised central area with loose white rather short mycelium; margin (fimbria) of the growing colony 2, 3 or 4 mm. wide, in color blue green C.d.C. 371, 372 at margin, 359, 360 within, becoming very dark shades such as 250, 224 in age; reverse in pale to brighter orange yellow tints such as C.d.C. 196, 146, 171, drops small, uncolored appearing in the marginal area; odor strongly fetid.



Conidiophores commonly varying from 300 to 600 $\mu$  long, 4 to 5 or even 6 $\mu$  in diameter, straight or slightly flexuous unbranched or rarely with one short branch figured as perpendicular, with walls sparingly asperulate; branches two, three or four in numbers commonly 20 to 30 $\mu$  long enlarging from base to the capitate apex with a range of 3 to 5 $\mu$ , with walls somewhat rough; metulae enlarging slightly from base to apex, 12 to 14 by 2.5 to 3.5 enlarging to 4 or 5 $\mu$ , usually in groups of 4 or 5; sterigmata straight, rather long, cylindrical 10 to 11 by 2.3 to 2.8, commonly 5 to 8 in the verticil; conidia smooth, subglobose, or occasionally ovate, 2.5 to 3.5 more rarely 4 $\mu$ , showing connections between the conidia and the chain.

Habitat: Species isolated from earth in pine woods at 8 to 10 cm. depth, in the forest known as "Puszcza Bialowieska." The designation of this form as new was concurred in by Biourge who suggested that it should also belong to a new series although Zaleski puts it in "*Euconcentrica classica*." Our notes on Zaleski's form follow: Type strain growing better at 20°C., than at 30°C. Colonies upon Czapek's solution agar about 20°C., slowly spreading, azonate or tardily and indistinctly zonate, with fasciculation of conidiophores evident at the margin and in radial section of the colony (more pronouncedly seen on wort agar plates) forming a mass varying from 100 $\mu$  deep at the thinnest white margin of the growing colony to 800 $\mu$  deep in the central area; bluish green to grayish olive and finally mouse gray; reverse colorless to dull yellowish gray, slowly becoming (several weeks) shades of brownish vinaceous at margin; drops colorless, well distributed over the colony; odor, faint or indefinite; conidiophores 100 to 600 or 700 $\mu$  by 4 $\mu$ , with walls pitted or roughened; penicilli up to 50 $\mu$  long or longer, consisting of a main axis, one or more branches with metulae and sterigmata at different levels, and chains of conidia forming more or less definite columns which may or may not break up in age into tangled masses; conidia elliptical 3.5 to 5 by 2.4 to 3 $\mu$  or less commonly subglobose 4 $\mu$  in long axis, separating in alcoholic mounts.

Culture no. 5010.33 received as type from Baarn in July, 1928, appears to be correctly identified and to be closely related to *P. patulum* of Bainier.

336. *P. brunneo-violaceum* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 145-147; Col. Pl. II, Cart. 2; Pl. IV, fig. 21. 1923.

Colonies on wort gelatine scarcely or not sharply zonate but included in zonate series, semifloccose or velvety with a felt of hyphae at base, quickly producing conidial areas bluish (C.d.C. 422) to blue green (C.d. C. 367) then green, finally violet-brown, with white margin 1 to 2 mm.

broad while growing; coremia none, or showing fascicles at the margin on Raulin-Lutz; reverse at first yellow then violet red; odor moldy; gelatine liquefied, yellow to brownish red; conidiophore  $3.5$  to  $4\mu$  in diameter; penicillus  $40$  to  $60\mu$  long with all walls smooth and parts falling away when ripe, figured as a main stalk with or without a single appressed branch, then verticils of metulae closely packed, enlarged at apex and bearing rather coarse sterigmata; branches  $18$  to  $20$  by  $2.2$  to  $3.8\mu$ ; metulae  $7.5$  to  $17\mu$  commonly  $12$  to  $14\mu$  by  $2.8$  to  $3.8\mu$  usually in threes, swollen at apex; sterigmata  $9$  to  $13$  by  $3$  to  $4\mu$ , in verticils of  $2$  to  $4$ , quickly falling away when ripe; conidia at first elliptical  $3.5$  to  $4.5$  by  $2.5$  to  $3.2\mu$ , soon globose  $4.5\mu$ , deciduous.

Biourge's no. 2 (our no. 4733.23) was received from him in September, 1927. Our notes follow: Colonies in Czapek's solution agar forming close felts about  $500\mu$  deep, more or less definitely zonate in the marginal areas, with surface unevenly fibrous tufted from small ascending to erect fascicles or ropes of conidiophores well distributed throughout the colonies, in color in dark yellowish green shades such as celandine or artemisia green (Ridgway XLVII); reverse in yellow to orange red shades such as Japan rose, testaceous, buff-pink in Ridgway XXVIII; agar partly colored especially beneath the colonies; conidiophores up to  $500\mu$  by  $3$  to  $4\mu$ , with walls pitted; penicilli, asymmetrically branched, with  $2$ ,  $3$  or  $4$  stages of branching, and having metulae and sterigmata caducous; metulae at different levels and differing considerably in length; sterigmata  $8$  to  $13\mu$  long, sometimes up to  $16\mu$ , with chains of conidia tangled; conidia from  $3$  by  $2\mu$ ,  $3.5$  by  $2.5\mu$  to  $4$  by  $3\mu$ .

When grown on wort slants aerial mycelium is more abundant and yellowish orange in color.

*Sub-section 5. Fasciculata-Coremiella.* Fasciculation fairly general, fascicles or coremia crowded, occasionally with longer prominent and branching forms; reverse and substratum mostly yellow to orange or reddish. Occasional cultures of *P. expansum* and its allies gives the appearance described here. Separation of some of these forms from others placed in Sub-section 5 is doubtful. In these cases, a sector taken from the culture with a sharp needle when examined along the cut edge (radius of the circular colony) has shown fasciculation under a  $10\times$  magnifier.

340. *P. corymbiferum* Westling. Arkiv för Botanik 11, pp. 56, 92-95; figs. 16, 58. 1911.

Colonies in prune gelatine, with white margin, mealy or granular white, and somewhat wooly then a central area blue-green (C.d.C 397, 353, 358,

363) with surface growth consisting principally of conidiophores, aggregated into small crowded coremia up to 5 mm. in height upon prune gelatine, with feather-like branching above, several coremia often arising from a common base, in the central areas and single coremia toward the margin; gelatine slowly and partly liquefied, beginning at seven to eight days and fairly complete in fifteen days; reverse yellow to orange; vegetative hyphae 2.4 to 6 or  $7\mu$ , with calcium oxalate sphaerocrystals abundant and some columnar crystals scattered among them; odor none; conidiophores with walls rough or almost smooth at times, 45 to  $700\mu$  long, by 4.2 to  $6\mu$ ; penicillus 40 to  $120\mu$  long, figured as main stalk and appressed clavate branch with verticils of metulae and sterigmata at a uniform level, symmetrical; metulae 12 to 16 by 3.2 to  $4.5\mu$ , with walls rough, or smooth; sterigmata 8 to 9.6 by  $3\mu$ ; conidia globose, smooth 2.6 to  $3.2\mu$ , occasionally 3.8 to  $4\mu$ .

Species found on orange juice and later on a lily bulb which showed decay and masses of green spores between the scales (see also Sorauer Handbuch der Pflanzenkrankheiten). In culture this species resembled *P. cyclopium*. It grew poorly at  $30^{\circ}$  to  $31^{\circ}\text{C}.$ ; It failed to grow on tannin solution but grew well on malt-extract-gelatin, prune gelatin, potato and bread, producing coremia up to 1 cm. in height.

Westling sent us his type culture (no. 2537) which in preliminary cultures appeared to belong with *P. granulatum* Bainier which we already had in culture. Unfortunately the culture was lost before adequate studies could be made. Biourge's culture (our no. 4733.43) does not belong to but is nearly related to *P. cyclopium*, hence we believe it incorrectly identified.

In January, 1929, we received no. 5034.64 from Nobel's Explosions Company in Scotland. This culture produced the two or three striking characters given by Westling. The full notes read: Colonies upon Czapek's solution agar, at first azonate, developing more or less zonation in a growing period of about 2 weeks, plane, about 300 to  $500\mu$  deep, fasciculation seen at margin and seen throughout when bits are torn from central areas and viewed in section with the handlens, with margin broad almost arachnoid in appearance, with areas of yellow to orange (fascicles orange) as it spreads over the substratum, passing to shades of bluish green with ripening conidia and to dark olive gray in age; reverse in shades of deep orange, reddish orange, brown or almost black, with reddish tinge sometimes in the thin edge of colonies on slanted agar; drops none at first, then orange to deep red; odor strong approaching that of *P. biforme* (suggestive of *Actinomyces*); conidiophores varying from short

to very long, 3, 4 or  $5\mu$  in diameter with walls granular; penicilli consisting of main axis and branches at 2 or 3 levels with metulae and sterigmata at various levels, arranged into a fairly compact brush or broom, 70 to  $80\mu$  long; conidia mostly about  $3\mu$ , some less, a few 3.5 to  $4\mu$ .

When no. 5034 Ad. 64 was received and colonies developed the resemblance to Westling's description became early evident.

This clearly excluded Biourge's strain from the species. When fitted into the classification however it was found to be nearly related to the strains from bulbs which had already been assigned to *P. hirsutum* Dierckx fide Biourge whose type however has not been seen. The strains are not identical but so nearly related that we would not be inclined to describe them separately. Dierckx's published description is worthless; combined with Biourge's notes (1923) our identification is believed to be correct to the series, perhaps not to the strain. Westling's description is however adequate and antedates Biourge. We are therefore inclined to call the series *P. corymbiferum* leaving *P. hirsutum* as possibly a species, more probably not more than a variety.

341. *P. hirsutum* Dierckx. Soc. Scientifique Bruxelles 25: 89. 1901.

In Biourge Monogr. La Cellule 33: fasc. 1, pp. 157-159; Col Pl. II, Cart. 186; Pl. III, fig. 18. 1923.

Biourge gives: Colonies on wort gelatine hemizonate, zonate especially at margin and in drier areas of medium, olive green, later brown, coremial tubercles showing in center on Raulin; overgrowth of mycelium predominates in age; reverse cream to yellow or tan; gelatine liquefied, with liquid yellow; drops yellow or orange to reddish or ruby, abundant, large, very conspicuous; conidiophore 4 to  $6\mu$  in diameter, with walls at first smooth (!?); penicillus about  $45\mu$  long, figured as a main axis often enlarged above the last septum into an ampulla 8 to  $10\mu$  in diameter, with appressed branches and metulae crowded both on the branches and on the apex of the ampulla; branches 17 by 3 to  $5\mu$  in two or threes; metulae 9 to 13 by 3.5 to  $5.5\mu$ , not inflated at the apex! sterigmata 7 to 11 by  $4\mu$ , in verticils of 2 to 4; conidia globose 3 to  $4\mu$ .

The culture received from Biourge as no. 186 (our no. 4733.71) failed to satisfy the description but proved to belong with *P. brevi-compactum*. Later a culture (no. 4979.2) found upon Spanish iris from California by F. Weiss of the Bureau of Plant Industry, satisfied the description given by Biourge fairly well. Our own notes follow (fig. 64): Colonies upon Czapek's solution agar, spreading, with white margin rather broad,

azonate (or hemizonate) forming a mass nearly  $1000\mu$  deep in the older areas, obviously fasciculate at the margin as seen with the hand lens and with fasciculation of the conidiophores evident throughout in microscopic preparations; conidial areas between bluish glaucous and dull glaucous blue (Ridgway XLII) with occasional yellowish more or less sterile areas among the blue green areas, reverse yellow to dirty orange shades passing to dark red shades in age; characterized by an abundance of large yellow to amber, even almost ruby drops scattered freely over the whole surface of the growing colony; odor slight; conidiophores with walls pitted (or rough), 4 to  $5\mu$  in diameter and up to several hundred microns long, colorless in young areas, yellow or an orange shade in older areas, simple or produced in fascicles whose yellow or orange basal portions are commonly visible with the hand lens in radial



FIG. 64. *P. hirsutum* Dierckx: Diagrammatic radial section (magnified by  $10\mu$ ); the figure over-emphasizes the fasciculation of the central area since the masses of the penicilli are so crowded as to form a dense green cover but the fasciculation of the stalks can be seen on the edge of the cut section.

sections of the central areas of colonies; penicilli fairly compact with conidial chains in groups commonly closely parallel to form one to three or loosely columnar masses 100 to  $200\mu$  long; branches 1 to 2 or 3, appressed or diverging at the tip, up to  $30\mu$  long; metulae about 10 to  $12\mu$  long in close groups; sterigmata about  $10\mu$  long; conidia mostly about  $3\mu$  in diameter.

Another culture (no. 4979.3) by Dr. Weiss from hyacinth bulbs imported from Holland is practically identical with 4979.2. Two cultures (no. 4811, x and z) received from Miss Clara A. Pratt of London, England, appear to belong with these forms to make a species aggregate of a series of strains with essential features in common. Which, if any, of these strains forms Biourge's type can not be determined, but the group identification is satisfactory.

342. *P. clavigerum* Demelius. Verhandl. Zool.-Bot. Gesellsch. Wien 72: 74-75, fig. 4, (1922) 1923.

Colonies in prune gelatine gray green (C.d.C. 371-346, 373-347; then blue green 362, 363, 368 or green 339, 350) slowly (1 month) and partly liquefying gelatine, characterized by abundant clear gray coremia 2 to 3 mm. in height around a blue green central area; reverse yellow; odor none; conidiophores mostly aggregated into coremia, up to 3 mm. by 4 to 4.6 $\mu$ , with walls smooth; coremia characteristically clavate, with stalk and head, not or rarely branching below the head in a feathery manner, with stalks yellow under the microscope; penicillus figured as main axis, and one fairly long branch bearing metulae few in the verticil, 40 to 60 $\mu$  long; metulae 9.6 to 14.5 by 3 to 3.6 $\mu$ , commonly 2 in the verticil; sterigmata 6 to 9.6 $\mu$  by 1.5 to 2.4 $\mu$ , commonly 3 to 5 in the verticil; conidia ellipsoid 2.3 to 3.6 by 2.3 to 3 $\mu$ .

Species found as gray yellow coremia upon a chestnut (*Castanea vesca*), 1916, in the Vienna market, but presumably from Hungary.

Demelius in her Latin diagnosis gives the conidiophores as up to 3 mm. long by 4 to 4.6 $\mu$  in diameter. In her German notes she describes the coremia as 2 to 3 mm. high, mostly branching at the apex to form a "head" but only rarely branching lower down to give a feathery aspect to the coremium. We have not seen a species which satisfies the details of this description.

343. *P. divergens* Bainier and Sartory. Bul. Soc. Mycol. France 28, 270-276, Pl. XIII, fig. 3-6. 1912. Reproduction of figure as fig. 65.

Colonies on licorice sticks at first bluish green (C.d.C. 371-372), then yellow green (C.d.C. 263, 264), characterized by abundant coremia up to 5 mm. in height, large at the yellow base and splitting into diverging bundles from which individual conidiophores diverge evenly over the upper third of the length; single conidiophores diverging from the column or coremium, about 5 $\mu$  in diameter, with walls minutely granular, with appressed branches at 1 level or more becoming 3-5 metulae, and small verticils of sterigmata; conidia slight oval or subglobose about 3 $\mu$  in long axis.

Species found associated with *P. granulatum* which it resembled closely; cultures grew best at 25° to 27°C. and produced a red pigment soluble in alcohol, acetone, ether, etc., little changed by acid but changed to blue in alkali. It differs from the related species by the aggregation of coremia at the base and their uniform divergence at the apex, by less

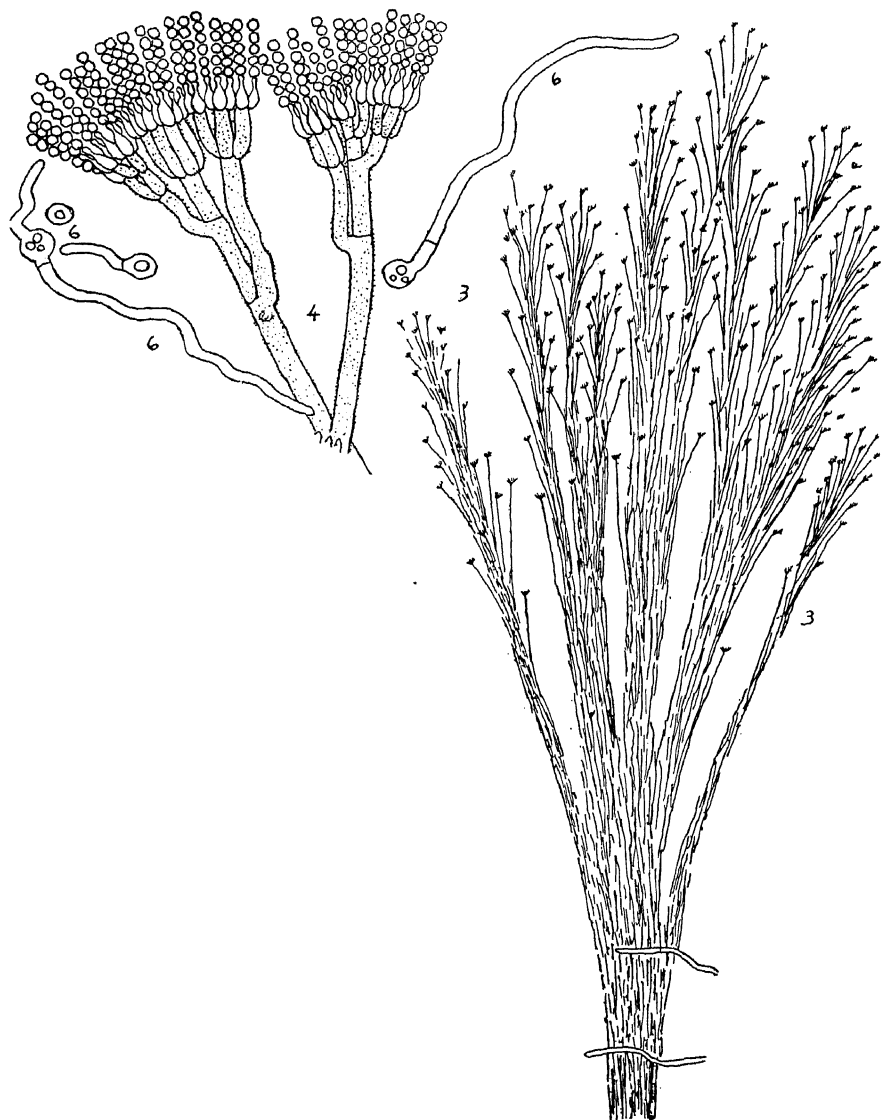


FIG. 65. Bainier's figure of *P. divergens*

prominent granulations of the stalk walls than those in *P. granulatum*. Our no. 4641 received from Dr. R. Thaxter, complies with this description by showing close relations to *P. granulatum* but producing prominent coremia spraying out beautifully at the tips. This culture bore a MS. name—*Coremium isarioides*. Biourge suggests synonymy with *P. aeruginosum* Dierckx but we have already decided that Dierckx was justified in placing that species with *P. italicum*.

344. *P. granulatum* Bainier. Bul. Soc. Mycol. France 21: 126–127; Pl. 11, fig. 2–7. 1905. Compare figs. 66, 67.

Colonies on licorice sticks, characterized by Stysanus-like coremia, consisting of a fascicle of conidiophores up to 3 mm long, spreading to form individual fruit masses from the middle upwards, at first white, with ripening conidia yellow green; conidiophores with walls finely granular or echinulate, arising as simple stalks among the coremia or as units in the coremia; penicillus figured as consisting of 1 or 2 branches, then metulae sterigmata and chains of conidia, all cell walls echinulate, no details of measurement given; sterigmata falling away in age; conidia averaging 2.6 by 2.1 $\mu$ , globose or elliptical retain their viability for one year or more.

Species found on pieces of oakwood. Bainier's type culture was given to the author by Gueguen when Bainier's laboratory was visited in 1905 and formed the basis of the description in Thom 1910, pp. 44–45, fig. 11, emended as follows: Colonies upon plain gelatin and potato or bean agar yellowish green to gray or grayish brown, superficially composed of crowded small coremia 1 to 3 mm. in height mixed with floccose hyphae and separate conidiophores, spreading indeterminately upon the substratum; reverse reddish orange ((approaching "fulvous") aerial hyphae delicately granular or spinulose, conidiophores 4–4.5 $\mu$ , in diameter, short or very long, either separate or, mostly, massed into very short, crowded coremia; penicilli usually 100 to 200 $\mu$  in length, once or twice verticillate, with many sterigmata 9 by 2.5 $\mu$ , and long, loosely divergent chains of conidia; conidia at first cylindrical, then elliptical to globose, about 2.5 to 3 by 3 to 3.5 or 3 $\mu$  in diameter, yellowish green, granular, remaining in long chains in fluid mounts. Colonies do not liquefy gelatin, litmus reaction slowly alkaline.

The delicately granular or spinulose hyphae were noted and figured by Bainier as a distinctive character but are much more common than Bainier then supposed them to be but the whole aspect of the colony is sufficiently definite to bring together various strains differing in detail



into a series of related species or varieties. Our transfer of the type no. 9, was lost. Fonseca in his transfers from the Bainier Collection sent us our no. 4640.446 which is nearly if not identical with it.

345. *P. granulatatum*. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 159-161; Col. Pl. II, Cart. 13; Pl. III, fig. 17. 1923. See also Biourge La Cellule 36: 454. 1925.

Colonies on wort gelatine hemizonate, forming a mass 1 mm. or more deep, more or less loose, with simple conidiophores mixed with coremia,

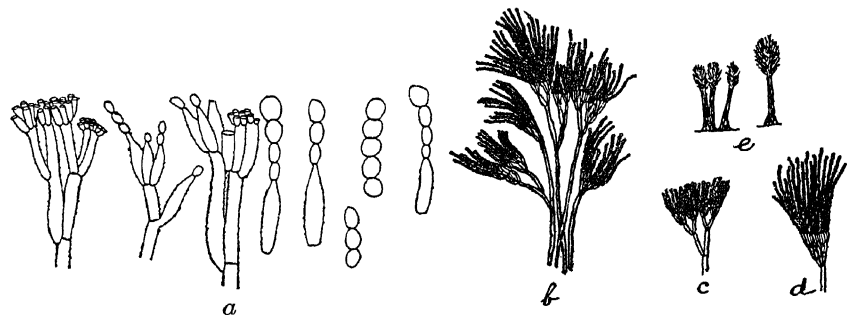


FIG. 66. *P. granulatatum* Bainier: Detail of penicillus and habit sketches with incipient fascicles at *b*, and large fascicles at *e*.

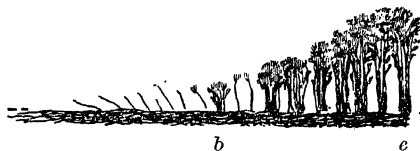


FIG. 67. *P. granulatatum*: Diagrammatic radial section

checked and variegated in color with conidial areas at first very bluish green, then blue green, with broad white margin, 2 mm. submerged, 2 mm. aerial; gelatine liquefied and colonies wrinkled irregular boat-like in age; reverse yellow to orange; odor none; drops yellow to red, commonly orange red; conidiophore 4 to 5 $\mu$  in diameter, with walls rough; penicillus about 50 $\mu$  long, figured as main axis and 1 branch, or in the sketch 1 or more distant and diverging branches, with closely packed verticils of metulae; branches about 30 by 3.5 to 4.5 $\mu$ , with walls rough; metulae 11 to 13 or even 17 by 3.5 to 5.5 $\mu$ , in verticils of 3 to 5; sterig-

mata 11 to 14 by 3 to  $4\mu$ , in verticils of 3 to 5; conidia ovate or lemon-shaped, up to 5 or  $5.5\mu$  by  $3.5\mu$  to 4, or globose  $3.5\mu$ .

Biourge's no. 13 was not received by us but was acknowledged not to be *P. granulatum* after he had received Thaxter's culture. Description puts it near *P. granulatum* and *P. corymbiferum* but hardly identical with either of them. When again found it will probably prove to belong in the section as a species or a variety of one of these species.

346. *P. nigrescens* Junghuhn. Bataviaasch Genootschape Verhandelungen V. 17. Praem. Flor. Crypt. Java. 1839.

Synonym: *Coremium nigrescens* (Jungh.) Penz. Fungi Agron. No. 132, Sacc. Syll. 4: 583; list in Syll. XI, p. 238.

Junghuhn figured his species as coremia scarcely  $\frac{1}{2}$  line (1 mm. in Saccardo) in height, scattered in groups over a piece of bark; with stipes composed of clustered or fascicled hyphae, spreading at the apex into a floccose brown head said to be composed of penicillate conidial fructifications; the stipe was (spadiceo-nigrescens) chestnut toward black at base; the conidia large and globose.

The type specimen was collected upon a Citrus trunk in April in Java. The species has not been reported since but the figures might satisfy some species like *P. granulatum* while the darker colors might be accounted for by the nature of the substratum and the age of the specimens when examined.

*Sub-section 6. Coremia.* Species with penicillate fruits diverging from a head-like mass borne upon conidiophores more or less closely aggregated into a stalk, while the production of simple conidiophores is suppressed or inconspicuous.

Link's genus *Coremium* was defined as a fruiting head borne upon a composite stalk composed of aggregated conidiophores. Thom, in 1910, expressed the belief that *C. glaucum* of Link's original series was the coremium form of *P. expansum* as universally found upon rotting apples in storage. Used as a subsection here, we have collected such forms as *P. claviforme* Bainier, whose type culture we have and find identical with *Coremium silvaticum* Wehmer, whose type culture is also in our collection. The coremia of *P. expansum* upon rotting apples, pears, grapes, and highly sweetened substances in nature would be placed here if we did not know their other relationship. *P. briardi* falls here by description and Metarrhizium as seen in culture produces forms with this general morphology. Compare the coremiform members of the Biveriticillata (Chapter XX).

Wehmer chose to revive the genus *Coremium* for his *C. silvaticum*. It is doubtful if anything would be gained by separating certain of these coremigenous forms which do not have the simple penicillate form also as we know them, from others which only occasionally appear as coremia without the simple penicilli. We have, therefore, included them here with no changes of nomenclature. Until some one really studies them all in a comparative manner, it is hardly profitable to offer new nomenclature but rather to record the observation that an organism unrecognized as met in culture may be found described among either group.

350. *P. claviforme* Bainier, Bul. Soc. Mycol. France 21: 127, Plate XI, figs. 8-11, 1905; Saccardo Syll. fung. 18: 520; Thom U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118: 44, fig. 10, 1910. Compare our figure 68.

Synonym: *Coremium claviforme* (Bainier) Peck (C.H.) in New York State Museum Bul. 131, p. 16, 1909, from a culture contributed by Thom; also *Coremium silvaticum* Wehmer Ber. deut. botan. Ges. 31 (5): 373-384, 1914; *P. silvaticum* (Wehmer) Biourge Monogr. La Cellule 33: fasc. 1, p. 105, 1923; *P. silvaticum* (Wehmer) Gäumann Verh. Morph. Pilze fig. 113, p. 177, 1926, in which change of name is attached to figure only; *Coremium vulgare* Corda Prachtflora part not all figures Pl. XXV may be this species.

Colonies on Czapek's solution agar, white or gray with more or less loose floccose hyphae bearing scattered inconspicuous simple penicillate conidial masses among the bases of conspicuous coremia with stalks compact, fibrous, white to flesh color or rose, up to 1 to 2 cm. long, simple or branched bearing well differentiated heads olive green in color and producing long conidial chains massed into columns often up to 1 to 3 mm. long and splitting variously with increasing length; reverse brown in age especially at bases of the stalks; odor strong, penetrating; drops abundant during the growing period, colorless; simple penicilli sparingly branched bearing verticils of few sterigmata 9 to 10 by  $2\mu$ ; heads composed of complex hymenium-like masses covered with sterigmata, crowded, radiating from the surface; conidia elliptical 4 to 4.6 by 3 to  $3.3\mu$ .

Bainier's type was obtained in his laboratory in 1905, but was lost; a duplicate received in the Bainier collection bears our no. 4640.441; Biourge's culture is our no. 4733.391; another culture was collected in

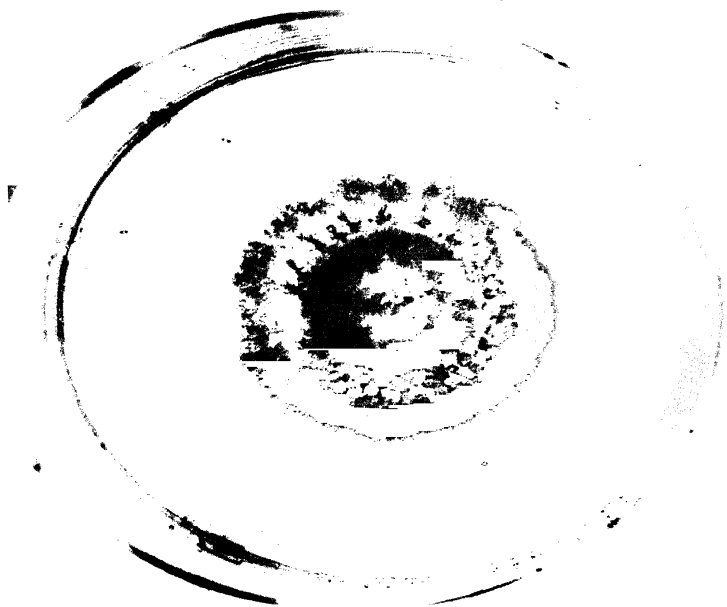


FIG. 68. *P. clariforme*: Photograph of colony showing a zone or ring of coremia



New York State by Reddick; *Coremium silvaticum* Wehmer as received from Wehmer is certainly the same species and may be a better name for it.

Killian and Lagarde discussed an unnamed species which resembles *P. claviforme* in figures and description. It was obtained from the intestine of a salamander. The coremia became 5 to 10 mm. high; heads 2 to 4 mm. long; sterigmata 8 to 10 $\mu$  in length; conidia ovoid, hyaline, 5 to 6 by 2.5 to 3 $\mu$ . These conidia are larger than we find in *P. claviforme*.

Wehmer also cited as possible synonyms which are listed here without review: *C. glaucum* var *finicola* Marchal, 1895; *C. cinereo-album* (Bon.) Sacc., 1892.

351. *P. claviforme* Bainier, var. *minus* Biourge, La Cellule 36: 454. 1925.

Biourge assigns this varietal name without description to a culture sent to him by H. G. Derx of Delft. He states that it bears his new no. 5. We have not seen it.

352. *P. briardi* Vuillemin, Bul. Soc. Mycol. France 20: 218-221; Pl. 11, fig. 9-10. 1904.

= ? *Isaria truncata*? Briard, Florule Cryptogamique de l'Aube. Troyes 1888, p. 457, no. 1672.

Not *Isaria truncata* Persoon, syn., p. 687; Sacc. Syll. IV. p. 584.

According to Briard: Coremia caespitose, about 2 cm. in height, branched, thickened, and cut or split at the apex, mealy; conidia 6 to 10 by 2 $\mu$ , oval to cylindrical, hyaline, unseptate.

In his material Vuillemin reports: Conidial apparatus more or less penicillately branched resembling *M. anisopliae*; conidia hyaline, more or less variable in size, about 6.5 by 2.2 to 2.8 $\mu$ , somewhat enlarged and rounded at the ends, suggesting the green muscardine (*Metarrhizium anisopliae* (but uncolored). Species isolated from an *Isaria*-like stalk borne upon a buried chrysalis of *Agrotis segetum*; this structure reproduced the description given by Briard for *Isaria truncata*. Conidia from this species have been used to infect insects successfully. Perithecia were not found.

Such organisms as this may be justly excluded from *Penicillium* and perhaps had better be regarded as species of *Isaria* until their real relationship is determined.

Von Höhnelt on p. 133 of his "Fragmente" refers to this organism as *P. briandii* Vuillemin.

353. *P. anisopliae* (Metchnikoff) Vuillemin, Bul. Soc. Mycol. France 20: 214-222; Pl. 11: 1-8. 1904.

More correctly *Metarrhizium anisopliae*.

Vuillemin cites: Giard, A., Bul. Scient. de la France et de la Belgique 1889, XX, p. 120-136, in a critical analysis of Krassilstschik's *De insectorum morbis qui fungis parasites efficientur* (place of publication not given) quoted Krassilstschik's statement that Metchnikoff in his Russian article (title translated) *Maladie des Hannetons du Blé*, Odessa 1879, used the name *Entomophthora anisopliae* for this species before he used the name *Isaria destructor* for it. Vuillemin therefore accepts the specific name first used, and transfers it to *Penicillium* as noted above. Delacroix, in Bul. Soc. Mycol. France 9: 260-264; Pl. XIV, 1893, decided that this species was not an *Isaria* and names it *Oospora destructor*.

Vuillemin reports in his cultures chlamydospores 7 to 9 $\mu$  in long axis, with thick walls and more or less globose, developed in the mycelium, conidia about 10.5 by 2.5 $\mu$ .

*Metarrhizium* is introduced here because investigators dealing with molds especially from soil occasionally encounter strains of an organism which apparently belongs with this genus but in ordinary culture media assumes much the aspect of a *Penicillium*. Colonies as seen upon Czapek's solution agar spread broadly in the substratum producing irregular conidial areas in which there is sharp differentiation of stalk and head. The whole mass may vary from a fraction of a millimeter in diameter to several millimeters in diameter, or form an irregular area up to one or more centimeters across, olive green in color and consisting of massed conidial chains borne upon penicillate branching systems arising either from single conidiophores aggregated into stalks or from branches from the massed tips of such stalks.

Such colonies give no hint of origin in parasitism upon insects. They are found often enough to lead to a doubt whether some of them may not conduct an independent existence as saprophytes with more or less omnivorous habits.

354. *P. cicadinum* v. Höhnelt. Sitzungsber. der Kaiserl. Akad. Wissensch. Wien Mathem.-Naturw. Klasse 118, Abt. 1 (pp. 131-133 in reprint). 1909.

On large singing cicadas white, then clear blue green and finally olive green from conidial mass; fruiting hyphae branched, bushy, colorless, smooth, 2 to 3 $\mu$  in diam. at upper end of sterigma; chains of conidia

more than  $100\mu$  in length, parallel, massed; conidia rod-shaped, rounded at ends, 5 to 6 rarely  $7\mu$  by 1.5 to  $2\mu$ , (germinating?).

v. Höhnelt states that this form may be *Oospora* rather than *Penicillium* because the branching of the fruiting hyphae is not typically penicillate.

Pathogenic to cicadas.

Slide in v. Höhnelt collection, Farlow Herbarium, labeled H.410, Buitenzorg, 1908, shows conidia yellowish green, of the *Metarrhizium* type, rod-shaped as described by v. Höhnelt, with rounded ends, vacuolate or nucleolate, massed in parallel rows. Tube containing cicada with wings presenting a dark dull green dusty mass of mold. Herbarium sheet, 1907-8, Java, Tjibodas, with note giving spores 6 by  $1.6\mu$ , 5 to 6 by 1.5 to  $2\mu$ , 7 by  $1.6\mu$ ; chains  $160\mu$  in fascicles.

### 355. *Isaria* sp.

The miscellaneous student of penicillate molds will occasionally meet forms with erect or ascending columnar masses, commonly pointed at the apex and bearing *Penicillium*-like conidial masses variously distributed along its length. These observations are in harmony with Atkinson's studies of the germination of spores from *Isaria* sp. as collected upon insects in nature (which we have repeated). Identification of such species with forms already described is not possible in routine culture. The finding of such species in cultures from soil is not uncommon but we do not yet know whether the mycelium of these fungi maintains a place in the soil flora independently of its parasitism upon insects. Thus far the *Isaria*-like mass has been occasionally seen but the relations have not been followed further.



## CHAPTER XX

### THE BIVERTICILLATA-SYMMETRICA

Wehmer in 1914 and Thom in 1915 described the symmetrically biverticillate type of penicillus as the outstanding common character of a genetic series of *Penicillia*. Wehmer proposed to call this section of the genus, the Verticillatae, without designating it as a sub-genus. Biourge substitutes the name Biverticillium and calls the section a sub-genus. He gives *P. purpurogenum* Fleróff-Stoll as the first species and his discussion clearly indicates that the type of structure found in the "Luteum Purpurogenum Group" of Thom or the Verticillatae of Wehmer represents in a general way his idea of the sub-genus but he goes on to include *P. aurifluum* (*P. citrinum* Thom) *P. atramentosum* Thom, *P. flexuosum* Dale (See Chapter XVII), and *P. fellutanum* which certainly do not belong with *P. purpurogenum*. Later in his correspondence with Zaleski we find that he has included the whole series centering upon *P. echinatum* Dale. These species only superficially resemble the *P. luteum*-like forms in having a biverticillate penicillus, hence we have reluctantly been compelled to discard the idea of a sub-genus Biverticillium and use the Latin adjective expression Biverticillata-symmetrica for the division of the genus with symmetrically biverticillate penicilli combined with the presence of lanceolate sterigmata. Two series of species with penicilli commonly biverticillate but asymmetrical are thus excluded: *Velutina-divaricata* and *Brevi-compacta*.

Monoverticillate and more or less definitely biverticillate penicilli are occasionally present in all species and more or less characterize an occasional strain which otherwise clearly belongs outside the biverticillate group. The symmetrically biverticillate penicillus is, however, too characteristic a feature of this group to justify the use of the name for these individual forms in other series. The conidiophore of this group produces a terminal symmetrical whorl or verticil of usually several metulae bearing symmetrical verticils of sterigmata closely packed in the verticil. The main axis prolonged forms the central metula. Occasionally this prolongation of the main axis produces a secondary or superposed verticil of metulae. Less frequently secondary verticils of metulae arise from the tips of individual primary metulae (see fig. 69).

The typical sterigma of the biverticillate group is a slender tube tapering at its apex to a long acuminate point (see fig. 69) from which the conidia are cut off as proportionately long cylindrical segments.

The conidia are at first cylindrical, usually swell quickly in the center and tend to assume fusiform to elliptical shapes, in some species becoming tardily globose or nearly so. The cell wall may be smooth or more or less roughened as a varietal or species character.

The whole aspect of this penicillus is so characteristic that once well understood the large majority of the members of the group encountered are allocated to the group at once from examination under the lower magnifying objectives of the compound microscope.

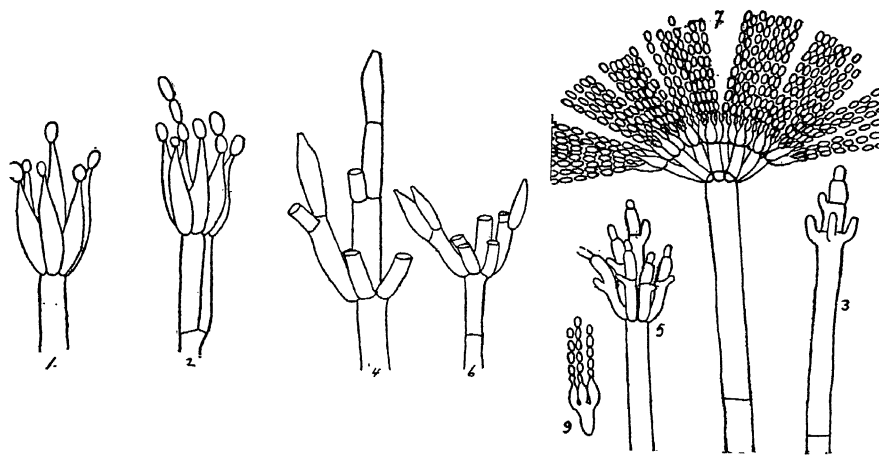


FIG. 69. The Biverticillate and symmetrical penicillus: 3, 5, 7 and 9 from Bainier's illustration of *P. olsoni* which are very diagrammatic 1, 2, 4, 6 from Thom 1910, *P. rugulosum*.

Species with the penicillus biverticillate and the sterigmata typically acuminate are readily shown to form a homogeneous series. In this same series, nearly all of the strains encountered produce yellow to orange or red colors in the mycelium and often also in the substratum. These colors and their transformation within the particular culture may develop slowly or quickly; they are intensified upon some media and more or less suppressed in others. In culture media containing fermentable sugars most of these species quickly produce an acid condition accompanied by yellow colors in reverse and often in the substratum.

With continued growth these colors may persist or change through shades of orange to rich deep red and this change follows changes in the hydrogen ion concentration of the medium and cell contents. In *P. duclauxi* the colors as diffused in the substratum are reversible with approximately the same relation to acid and alkali as phenolphthalein.

We have not determined what relationship, if any, may exist between forms such as these with colorless or bright colored hyphae and such a dematiaceous form as Pollacci described as *Haplographium De Bella Marengo*, yet figured with a biverticillate penicillus and reported as closely associated with *P. burci*. So many changes of color and structure arise as reactions to substrata that much combined field and culture work will be required to interpret such findings correctly.

Recognizing frankly that deceptive appearances and arbitrary allocations of species are both encountered, *Penicillia* biverticillate in appearance may be therefore separated again to assist the worker, as follows:

Species asymmetrically biverticillate.

- a. Metulae or branchlets and sterigmata divergent—*Lanata-divaricata*, Chapter XVII.
- b. Branch or branches, and metulae and sterigmata—closely aggregated at the base—*Brevi-compacta*. Chapter XV.

Species symmetrically biverticillate.

- a. Sterigmata acute pointed; conidia globose or subglobose—*Velutina-divaricata* Chapter XIV—some *Biverticillata-miscellanea*, nos. 420–430.
- b. Sterigmata acuminate or lanceolate; conidia mostly elliptical—*Biverticillata symmetrica* (strictly).

*Key to biverticillate and symmetrical species*

- I. Species reported as producing sclerotia or perithecia..... II
- I. Species not reported as producing sclerotia or perithecia..... X
- II. Sclerotia only, reported..... VIII d and h
- II. Perithecia and ascospores reported..... III
- III. Conidial areas not green..... IV
- III. Conidial areas showing some shades of green.... V
- IVa. Conidial areas avellaneous; conidia 3 to 3.5 by 2 to 2.5; ascigerous areas Indian red; ascospores 6.5 to 8.5 by 4 to 5μ..... *P. avellaneum* Thom and Turesson, 365.
- IVb. Conidial or non-ascigerous forms of same or nearly related species..... *P. capreolinum* Biourge, 366.

- V. Ascospores described.....VI  
 V. Reported as ascigerous but ascospores not described.....VII
- VI. Ascospores 4.8 by  $3.3\mu$ , transversely trico-state.....*P. luteum* Zukal, 367
- VIa. Ascospores about 4.6 by  $2.6\mu$  minutely spinulose.....*P. Wortmanni* Kloecker, 368.  
 Probable synonyms.....1. *P. luteum* in Thom 1910, 369.  
 2. *P. vermiculatum* Dangeard, 370.  
 3. *P. sulfureum* Sopp in Biourge, 371.
- VIb. Ascospores 3 by  $2.5\mu$ , thick walled; conidia 2 by  $1\mu$ .....*P. sacchari* Ray, 372.
- VIc. Ascospores 2.5 to 4 by 1.8 to  $2.8\mu$ , minutely spiny; conidia 2.5 to 4 by 1.8 to  $2.5\mu$ .....*P. spiculisporum* Lehman, 373.
- VIc. Ascospores 5 by  $3\mu$  yellow, smooth; conidia 3 by  $1.5\mu$ .....*P. aureum* Van Tieghem, 374.
- VIe. Ascospores about  $6\mu$  in long axis echinulate; conidial areas blue green; marginal areas canary yellow; conidia about  $4\mu$  in long axis.....*P. Petchii* Sartory and Bainier, 375.
- VII. Perithecia reported but ascospores not described.....VIII
- VIIIa. See VIa.....*P. vermiculatum* Dangeard, 370.
- VIIIb. Colonies sordid yellow; with scanty traces of greenish conidia.....*P. gilvum* Sopp, 376.
- VIIIc. Colonies with yellow green conidial areas upon insects; with sulphur yellow mycelium and "perithecia" in cultures.....*P. parasiticum* Sopp 377.
- VIIIc. Colonies green; reverse more or less yellow; conidia 3.5 to 3.8 by 3 to  $3.3\mu$  sclerotia yellow with greenish centers.....*P. kiliense* Weideman, 378.
- VIIIe. Colonies sulphur yellow with a greenish tinge; "perithecia" or sclerotia large, yellow green.....*P. citrinum* Sopp, 379.

- VIII f. Colonies green or yellow green from conidia 4 by 2.5 to 3 $\mu$ ; "perithecia" gray brown—not further described..... *P. virido-bruneum* Sopp, 380.
- VIII g. Colonies producing chalk-white mycelium and reddish to blood red perithecia in autumn, producing yellow-green conidial areas in spring..... *P. niveo-rubrum* Sopp, 381.
- VIII h. Sclerotia reported as occasional in..... 1. *P. hirsutum* Bainier and Sartory.  
2. See No. 426.  
3. in *P. olsoni* Bainier, 401.

For coremiform species not found here see Chapter XIX.

- X. Species producing erect, definite coremia bearing the conidia..... XI
- X. Erect coremia absent..... XV
- XI a. Erect crowded, commonly branching coremia with dark green conidia 3.5 to 4 by 2 to 2.5; reverse yellow to red..... *P. duclauxi* Delacroix, 385.
- XI b. Stalks of coremia yellow..... *P. bicolor* Fries, 386.
- XI c. Stalks of coremia yellow..... *Coremium citrinum* Corda, 386.
- XI d. Colonies in shades of blue green to gray green; reverse yellow; conidia given as 3 to 6 by 3 to 4 $\mu$ , slightly echinulate; coremia variously produced with stalks colorless, yellow or in red shades..... *P. Zukalii* Biourge, 387.

*Virido-lutea*

- XV. Colonies showing green conidial areas on a mycelium usually showing yellow shades (sometimes red in age)..... XVI
- XV. Colonies lacking the yellow to red mycelium... XXV
- XVI. Colonies superficially characterized by networks of hyphae and ropes of hyphae..... XVII.
- XVI. Ropiness reduced to inconspicuous amounts or absent..... XVIII.

*Funiculose series*

- XVIIa. Colonies deep green with yellow margin; conidia 2.5 to 4 by 2 to 3.2 $\mu$ .....*P. luteo-viride* Biourge, 390.
- XVIIb. Colonies olive grey with yellow green margin; conidia 2.5 to 3 by 2 to 2.5 $\mu$ .....*P. humicola* of Gilman and Abbott, 588.
- XVIIc. Colonies yellow-green, commonly in tufts; conidia 3 to 3.5 by 2.....*P. pinophilum* Hedgcock, 391.
- XVIId. Nearly related species.....*P. minio-luteum* Dierckx, 392.
- XVIIe. Colonies with margin yellow; reverse quickly red, becoming very deep red to almost black in age; conidia 3 to 4 by 2 to 3 $\mu$ .....*P. funiculosum* Thom, 393.
- XVIIf. Nearly related form: conidia 1 to 3 by 1 to 2 $\mu$ —specimen labeled.....*P. fastigiatum* by Geo. F. Atkinson, 394.
- XVIIg. Nearly related form: conidia given as 2.8 by 2.4 $\mu$ .....*P. africanum* Doeblet, 395.
- XVIIh. Colonies forming heavy bristly felts, showing three colors, conidial areas green, red areas in the older parts and yellow areas at the margin; conidia 3 to 3.5 by 1.5 to 2 $\mu$ .....*P. islandicum* Sopp, 396.
- XVIIi. Colonies rather deeply floccose, with hyphae, ropes of hyphae and long conidiophores largely covered with yellow granules; conidia 3.5 to 4 by 2 to 2.5 $\mu$ .....*P. herquei* Bainier and Sartory, 397.
- XVIIj. Probably related to this species.....*P. aureum* Corda, 398.
- XVIIk. Probably related: "wonderful rose oil odor"....*P. lemoni* Sopp, 399.
- XVIIl. Colonies bluish green, with coarse conidiophores up to 2 mm. by 8 $\mu$ ; conidia 3 to 3.5 $\mu$  in long axis.....*P. elegans* Sopp, 400.
- XVIIm. Probably identical.....*P. Olsoni* Bainier and Sartory, 401.
- XVIII. Colonies with dense dark green conidial areas, more or less bordered, mixed with, or overgrown by yellow to orange hyphae; reverse in orange shades occasionally almost red; conidia elliptical rough or mostly so.....*P. rugulosum* and allies. XIX.

- XVIIIa. Colonies with green conidial areas and reverse quickly and intensely purple red.....*P. purpurogenum* and allies. XX.
- XVIIIb. Colonies with green conidial areas, yellow sterile areas with predominantly yellow to orange rather than intense red in reverse.....Non-ascosporic *P. luteum* series XXI.
- XIXa. *P. rugulosum* Thom.....no. 405.
- XIXb. Morphology of *P. rugulosum* but with white marginal zone and colorless reverse.....*P. rugulosum* var. *atricolum* (Bainier) T., 406.
- XIXc. Closely related to *P. rugulosum*.....*P. chrysitis* Biourge, 407.
- XIXd. Morphology of *P. rugulosum* but forming deep and uneven masses of conidia; reverse colorless or in zones in redder shades of orange red (Corinthian red).....*P. crateriforme* Gilman and Abbott, 408.
- P. purpurogenum* series
- XXa. Colonies restrictedly growing, velvety, green to ochraceous red; reverse yellowish to red; conidia 3.4 by 2 $\mu$ , to 2.5 to 3 $\mu$ , produced in masses on sugar media.....*P. rubrum* Stoll, 409.
- XXb. Colonies zonate, deep green to dark olive green; blood red below.....*P. sanguineum* Sopp, 410.
- XXc. Colonies showing a mixture of yellow green and yellow to red colors.....*P. variabile* Sopp, 411.
- XXd. Colonies tending toward floccose felts with green conidial areas and deep red reverse.....*P. purpurogenum* Stoll, 412.
- XXe. Variety with the development of red to almost black sclerotial mass.....*P. purpurogenum* var. *rubrisclerotium* Thom, 413.
- XXI. Non-ascosporic members of the *P. luteum* series.
- XXIa. Mycelium in varying shades of yellow with more or less development of green conidia. Haplonts of.....*P. luteum* (looser sense).
- XXIb. Yellow area forming a marginal zone about the green conidial area.....*P. aureo-limbum* Zal., 415.

- XXIc. Yellowish green to blue green; conidia small.....*P. citricolum* B. and S. 416.
- XXId. Conidial areas dark gray to green to olive green upon a pale yellow mycelium; conidia 2 to 3 $\mu$  in long axis.....*P. luteum* of Sopp, 417.
- XXIe. Conidial areas in deep green to olive green shades interspersed with sulphur yellow sterile mycelium.....*P. sulfureum* Sopp, 418.
- XXIf. Colonies yellow green; conidia 3.4 by 1 $\mu$ .....*P. flavo-virens* Cooke and Massee.
- Miscellanea*
- XXV. Colonies divergent in structure of penicilli and without the yellow and red of the other biverticillate series or nearly so.....XXVI.
- XXVI. Colonies some shade of green.....XXVIII.
- XXVI. Colonies never green.....XXVII.
- XXVII. Colonies avellaneous; nonascosporic.....*P. avellaneum*. See 365.  
*P. capreolinum*. See 366.  
*P. gilvum* (?). See 376.
- XXVII. Colonies white.....*P. Braziliense* Thom, 420.
- XXVIII. Species known to us in culture or adequately described.....XXIX.
- XXIX. Colonies velvety.....XXX.
- XXIX. Colonies floccose.....XXXV.
- XXXa. Colonies upon Czapek with a thin colorless submerged mycelium, with a few pinhead clusters of green penicilli; odor strong; conidia 2 to 3 by 1.5 to 2 $\mu$ .....*P. Namylovskii* Zal., 421.
- XXXb. Colonies restrictedly growing, slowly developing green conidial areas; with broad fibrous or submerged margin; reverse uncolored; conidia 2.5 to 3 $\mu$  in long axis.....*P. tardum* (Bainier) Thom, 422.



- XXXc. Colonies gray green; figured to suggest this group; conidia 3.5 to 5 by 1.8 to 2.4 $\mu$ .....*Paecilomyces erectus* Demelius, 423.
- XXXV. Colonies floccose.....XXXVI.
- XXXVIa. Colonies felted fasciculate to funiculose in center, fimbriate velvety at margin, conidia up to 4 by 3 $\mu$ .....*P. brunneo-violaceum* no. 336.
- XXXVIb. Colonies loosely floccose, slowly developing pale bluish green conidial areas; penicilli very irregularly biverticillate; conidia 3.4 to 4.6 by 2.8 to 3.4.....*P. piscarium* Westling, 424.
- XXXVIc. Colonies cottony floccose yellowish green; penicillus from monoverticillate to irregularly biverticillate; conidia 2.5 to 3 by 2 $\mu$ .....*P. Miczynskii* Zal., 425.
- XXXVIc. Colonies gray green; conidia 3.5 to 4 by 2 to 3....*P. Rolfii* Thom, 426
- XXXVIId. Colonies floccose, gray green; penicilli regularly or irregularly biverticillate, conidia mostly 2.5 to 3 $\mu$ , at times 4 by 3 $\mu$ .....*P. matris-mae* Zal., 243.
- XXXVIe. Colonies floccose, green; conidia 2.7 to about 4 by 2 to 2.8 $\mu$ .....*P. lagerheimi* Westling, 427.
- XXXVII. Colonies loosely floccose; penicilli with long, tangled, divergent chains of conidia:
- Conidia 2 to 3 $\mu$  in long axis.....*P. desciscens* Oudemans, 428.
- Conidia 4 by 2 $\mu$ .....*P. exiguum* Bainier, 429.
- Conidia 2 $\mu$ .....*P. humicola* Oudemans. See Chap. XXV.
- Conidia 2.5 to 3 $\mu$ .....*P. hirsutum* Bainier and Sartory, 430.

## SECTIONS IN BIVERTICILLATA-SYMMETRICA

Numerous species already described are allocated to this division. Among them are several natural series of forms obviously closely related, and others whose affinities are thus far unsettled. For convenience in identification and comparison the available material is presented in four sections even though closely related strains may be separated at times by such division.

*Section 1. Ascogena.* The species producing perithecia and asco-

spores are put together as the *Ascogena*. One sclerotium producer is arbitrarily included. In making this aggregate *P. avellaneum* Thom and Turesson, and *P. spiculisporum* Lehman whose affinities are doubtful have been included with a truly homogeneous series including *P. luteum* and its allies so recently discussed by Derx and Biourge. The non-ascosporic members of the *P. luteum* series are arbitrarily put together in Section 3 for discussion even though continuation of Derx's studies will probably ultimately show forms described separately there to be haplonts of some ascosporic species to be determined later.

*Section 2. Coremigena.* Species like *P. duclauxi* Delacroix with abundant and erect coremia are sufficiently differentiated to form an arbitrary but useful section in such a classification as this.

*Section 3. Luteo-virida:* Colonies commonly showing green conidial areas on mycelium more or less yellow to red.

*Sub-section 3a. Funiculosa.* The species producing trailing and anastomosing ropes of hyphae have so much of morphology and biochemical reactions in common as to warrant segregation under the name of one of them as a series. Reverse and substratum are commonly yellow orange or red, in shades or successions characteristic of the strain or species.

*Sub-section 3b. Luteo-purpuregena.* Colonies with conidial areas some shade of green, commonly bordered or intermixed with areas yellow from hyphae covered with yellow granules which may become red in age; reverse and substratum yellow, to orange and often to red, differing in the intensity, rate of development and final shade of color attained.

*Section 4. Miscellanea.* The biverticillate type of penicillus is characteristic of a number of species part of which lack the typical sterigma and others lack the color reactions of *P. luteum* and its allies. These species have been aggregated into a sub-section of this group for taxonomic and descriptive purposes. The members of the section are not regarded as genetically related and some of them show little in common with the other biverticillate sections.

*Sub-section 1. Ascogena.* A series of ascogenous species known in culture from the work of Zukal, Wehmer, Klöcker, Dangeard, Ray, Thom, Derx, and Biourge, produce conidial apparatus of the *P. luteum* type in shades of green and yellow green. *P. avellaneum* of Thom and Turesson has a penicillus of related morphology. Instead of producing a hard sclerotium-like body which slowly develops into a perithecium over several weeks as described by Brefeld for *P. glaucum*, all of these forms produce soft ascogenous masses of hyphae with scarcely any

development of perithecial wall, in which ascospores develop quickly. This type of ascogenous mass lead Saccardo to transfer *P. luteum* to *Gymnoascus* (*G. luteus* (Zukal) Sacc. in the Sylloge XI, p. 437), perhaps correctly. However Lehman found his *P. spiculisporum* with a related and apparently reduced type of penicillus but with perithecia showing a soft, brittle but fairly definite wall. We have had several strains in culture so closely related to Lehman's organism that separation could only be based upon more complete studies than have yet been made. One of them (no. 4029.35) produces abundant white conidial areas with a penicillus approaching that of *P. avellaneum* but with perithecia resembling those of *P. spiculisporum*. Until more complete studies of perithecium formation in this group are published, we have left *P. wortmanni* and its allies to represent the yellow-green or *P. luteum* series of strains, *P. avellaneum*, to represent a series with conidial areas in avellaneous and related shades, and *P. spiculisporum* for those with conspicuously differentiated perithecia with well developed perithecial wall.

*Conidial areas not green or scarcely green.*

365. *P. avellaneum* Thom and Turesson. Mycologia 7:284-287, figs. 1, 2. 1915. See fig. 70.

Colonies upon Czapek's solution agar, broadly spreading, slightly floccose, in conidial areas becoming persistently avellaneous (Ridgway XL), producing perithecia slowly during a period of several weeks with the gradual development of aerial hyphae colored Indian-red in the perithecial areas; reverse and agar becoming Indian-red (Ridgway XXVII); conidiophores up to  $400\mu$  long by 3 to  $5\mu$  in diameter, bearing conidial fructifications up to  $200\mu$  long, composed of loosely parallel or tangled chains of conidia; fertile branches either a terminal crowded verticil of metulae 8 to 10 by  $3\mu$  bearing verticils of few sterigmata 8 to 9 by  $2\mu$ , or with branches more or less irregularly disposed over the terminal 10 to  $15\mu$  of the conidiophore; conidia ellipsoid to almost globose, 2 to  $2.5\mu$  by 3 to  $3.5\mu$ , smooth, swelling in germination to  $5\mu$  in diameter and producing a single tube.

Perithecia ellipsoid to globose  $300$  to  $600\mu$  in diameter, originating as an ascigerous mass surrounded by numerous swollen, very thick-walled cells, with the slow development of a peridium composed of thick-walled cells, 8 to  $12\mu$  in diameter in one or sometimes two layers; asci 9 to 10 by 12 to  $15\mu$ , 6 to 8-spored; ascospores ellipsoid, 4 to 5 by  $6.5$  to

8.5  $\mu$ , with walls thick, pitted or with the appearance of round, transparent spots.

The original culture, no. 4010.7, was isolated by Turesson from feces of a bear in the zoological garden at Seattle, Washington. As carried in the laboratory most cultures under this number quickly lost their ascospore producing power apparently confirming Derx in the conclusion that the mycelium of members of this whole group is unisexual, hence the loss of ascus production was due to the elimination of one of the necessary haplonts. Several cultures with the morphology of this species have been received from widely separate sources as follows: no. 4401, ascosporic from Guatemala City, Guatemala, on a cereal product; no. 4152.1 from Porto Rico; no. 4235.15 from China; no. 4733.15 from Biourge.

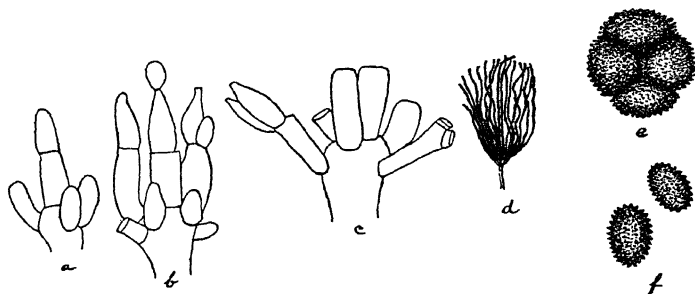


FIG. 70. *P. avellaneum* Thom and Turesson: a, b, c, detail of the branching of the penicillus, d sketch of penicillus and Turesson's figure of ascospores in the ascus e and separate f.

366. *P. capreolinum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 246; Col. Pl. XI, Cart. 311; Pl. XVII, fig. 102. 1923.

Colonies in wort gelatine velvety, in shades of orange yellow toward avellaneous, reverse brownish; conidiophore 2.5 to 4 $\mu$  in diameter, with walls rough; penicillus commonly about 25 $\mu$  long or lengthened to 65 $\mu$  or more by the occasional proliferation of metulae to form accessory verticils; metulae with walls echinulate, 9 to 15 by 2.5 to 3 $\mu$ , apex often enlarged, in groups or umbels of 3 to 8; sterigmata 10.5 to 12 by 2.5 to 3  $\mu$ , in verticils of 2 to 5, smooth; conidia more or less fusiform, 3.5 to 5.5 by 2 to 3 $\mu$ , slightly roughened.

Biourge no. 311 (our no. 4733.27) when grown on Czapek's solution agar produced colonies in shades of yellow orange toward avellaneous

and drab, but without true green; aerial hyphae commonly studded with granules; velvety 100 to 200 $\mu$  deep, with tuberculate center and overgrowths rising perhaps to 1 mm., with margin fairly bright yellow in the growing area, reverse deep orange to red orange to reddish brown; conidiophores and metulae more or less pitted; conidia mostly 3 to 4 by 2 to 2.5, with occasional individuals 6 to 7 in long axis.

The culture appears to be a non-ascosporic strain closely related to *P. avellaneum*.

*Conidial areas green or yellow green: Ascospores described.*

367. *P. luteum* Zukal. Sitzber. K. Akad. Wiss. (Vienna) Math. Naturw. kl. 98:521. 1888. See also Wehmer, Ber. deut. bot. Gesellsch. 2:499-516, Taf. 25, 1893; Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118:39, 1910; Biourge Monogr. 231-235; Derx, Bul. Soc. Mycol. France 41:375-381, 1925.

In Zukal's description the conidia were ellipsoid, gray green smooth 2.3 by 1.4  $\mu$ ; the ascospores 4.8 by 3.3, transversely tricostate; his other data we now recognize as locating his species in the biverticillate group, but not as placing it more closely.

Zukal undoubtedly was the first to describe an ascogenous biverticillate *Penicillium*. Wehmer followed Zukal in his nomenclature and described his cultures more fully. Thom carried his culture (no. 11) to Wehmer's laboratory and reported Wehmer's confirmation of the organism as *P. luteum*. Up to that time the probability of several nearly related ascosporic species was disregarded by all. Biourge questioned the identity of Wehmer's organism with that of Zukal, but did not clear up the difficulties. Derx with the ascosporic cultures accumulated by all previous workers in his hands, critically reexamined the ascosporic markings of the various strains and proposed the restriction of *P. luteum* to Wehmer's organism which produces ascospores with 3 or 4 cross-ridges which more or less completely reproduce Zukal's description. He identifies Thom's *P. luteum* (no. 11) with *P. vermiculatum* Dangeard, but since Biourge seems to have identified this same culture with material from the Carlsberg Laboratory which seems to be unquestionably *P. wortmanni* Klöcker, a name which antedates Dangeard's work, this name will be used for the no. 11 type of organism.

Two possible courses are open to the student of these species as molds, (1) Derx suggests limitation of the use of the name *P. luteum* to such ascosporic cultures as fully comply with the ascospore markings

described by Wehmer, since this is tentatively the nearest we can come to the organism of Zukal. Rigidly interpreted this would exclude the use of the name from non-ascosporic cultures presumably haplonts of this or some other ascosporic member of the series until the ascosporic form of such cultures could be determined; (2) since we need some means of identifying members of this related series, some of which are constantly encountered, the name *P. luteum* can be used as an aggregate followed by the limiting word, group. The second of these alternatives has been widely practiced up to the publication of Derx's paper and is continued here since Derx himself has only partially developed his scheme of nomenclature. In this section we have included forms which fall more or less naturally into a *P. luteum* aggregate, a *P. rugulosum* aggregate and a *P. purpurogenum* aggregate which may for convenience be called "series" in connection with these names.

368. *P. wortmanni* Klöcker. Compt. Rend. Laborat. Carlsberg. 6: p. 100. 1903. See Biourge, Monogr. La Cellule 33: fasc. 1, p. 244, 1923.

*Synonyms:* *Gymnoascus luteus* (Zukal) Sacc, Syll. XI, 437; *P. luteum* in Thom, 1910; *P. luteum* (Zukal) Thom in Biourge's Monogr., 243; *P. vermiculatum* Dangeard, q. v. fide Derx, Bul. Soc. Myc. France 41: 377, 1925.

Colonies at first white then sulphur yellow later at times orange; hyphae with cell walls covered with yellow granules giving the colony color; conidiophores and conidial areas produced irregularly, upon some media fairly abundantly, in others with such conidial areas few or suppressed; sterigmata 9 to 13  $\mu$  long; conidia ovoid or globose, mostly about 2  $\mu$  in long axis, but swelling greatly in germination and producing 1 or several tubes.

The ascus producing masses are yellow or reddish, enveloped in masses of hyphae crushing readily and rendering perithecial structure difficult to demonstrate; asci abundant in the crushed masses globose, ovoid, irregular, 8 to 13  $\mu$  in long axis, containing 8 spores; ascospores ovoid, about 4.6 by 2.6  $\mu$  marked over the surface with delicate points.

Species found by Klöcker in Danish soil.

Biourge (Monogr., p. 72) attributes his culture to Klöcker hence gives it the presumptive standing of *type*; Biourge's no. 401 (our no. 4733.126.1) may thus be regarded as certainly representing Klöcker's organism. In our own cultures there were minor differences in habit between no. 11 and no. 4733.126.1, but we are inclined to accept Biourge's

statement of identity. Ascosporic forms belonging here have been repeatedly isolated in our soil bacteriological plates.

Our no. 2424 (not now viable) was received from Westerdijk in 1910 as *P. wortmanni*. Without producing ascospores, this culture produced conidiophores manifestly belonging with the aggregate which Thom called the *P. luteum-purpureogenum* group, but did not suggest itself as identical with his no. 11 which Thom (Bul. 118) failed to recognize as *P. wortmanni*, rather identifying it in personal conference with Wehmer as *P. luteum* Zukal, ignoring uncertain observations of ascosporic markings which were always very difficult to determine. Biourge insists upon the identity of this form with Klöcker's species. Derx (1925), however, with the same culture (Thom's no. 11) in hand identifies it absolutely with *P. vermiculatum* Dangeard, ignoring Klöcker's species. If both these authors are correct, *P. wortmanni* certainly antedates *P. vermiculatum* hence must be attached to this species if *P. luteum* of Zukal with the tricostate markings on the ascospore is certainly identified as indicated in Derx's paper.

369. *P. luteum* (Zukal ?) Thom. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 243-244; Col. Pl. XI, Cart. 368; Pl. XVIII, fig. 103. 1923.

Synonym: *Certainly P. wortmanni* Klöcker (Biourge No. 401); probably not *P. luteum* Zukal.

Biourge's no. 368 (not received) was apparently Thom's no. 11 as sent to Biourge. Disregarding all reference to ascospores in his discussion, Biourge merely redescribes the organism sent him by Thom without material additions to that description. This name as attributed to Zukal and Thom should be dropped.

370. *P. vermiculatum* Dangeard. Le Botaniste 10: pp. 123-139. 1907.

Synonym: *P. wortmanni* Klöcker 1903; ascosporic *P. luteum* of Thom, 1910; fide Derx Bul. Soc. Mycol. France 41: 377, 1925.

Colonies on potato at first uncolored then slowly sulphur-yellow becoming more or less reddish in age, producing perithecia rapidly, usually in about eight days, with all stages from ripe asci in the center to beginnings at the periphery; on carrot the same changes but more rapid development; creeping hyphae not over 5 to 6 $\mu$  in diameter, producing very fine rhizoids, anastomosing freely, with cells 1-nucleate, conidial areas irregularly produced, sometimes wanting, bluish-gray in color; conidiophores slender, consisting of 2 to 3 cells; conidia elliptical

thick walled and delicately echinulate, 3 by  $2\mu$ , separating in mounts; perithecia abundant in all cultures, originating in 2 copulating clavate branches 1-celled, 1-nucleate and rich in granular protoplasm; the ascogone elongates, enlarges, becomes 8 to 16 nucleate, meanwhile the second branch "trophogone" from a different hypha, develops and forms several spirals about the ascogone, cuts off a terminal 1-nucleate cell which fuses with the ascogone by a pore, but without nuclear fusion; the contents of the ascogone now containing 32 to 64 nuclei break up into 2 nucleate cells each of which becomes an ascogenous filament developing among the trophogenic cells of the perithecial mass. Asci 4 to 6-spored; ascospores ellipsoid, spinulose but measurements not given.

Derx 1925, identified Thom's no. 11 as this species but Biourge had already shown that this same organism should be called *P. wortmanni* hence Dangeard's name should be dropped and Klöcker's retained. Derx's evidences as to such identity were not given.

371. *P. sulfureum* (?) Sopp. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 241-242; Col. Pl. VII, Cart. 195; Pl. XI, fig. 63. 1923.

Colonies on wort gelatine more or less velvety, olive, dark olive with yellow (sulphur yellow) margin, or whole colony yellow, coremia none; reverse orange to yellow (upon Raulin colonies greenish in reverse at first then orange); odor agreeable; conidiophore 3 to  $4\mu$  in diameter, often alternate toward the base, with all walls smooth; penicillus either a simple umbel or biverticillate form about 20 to  $30\mu$  long, or with 1 or more proliferating metulae to produce a double length 50 to  $65\mu$ ; branches rare; metulae 12 to 20 by 2 to  $4\mu$  in groups of 4 to 7, one or more of which usually produces a secondary verticil (umbel); sterigmata 11 to 15 by 2 to 2.5 in groups of 2 to 5; conidia more or less fusiform, 2.5 to 4.5 by 1.5 to  $2.5\mu$ , smooth, commonly apiculate.

Biourge's no. 195 (our no. 4733.120) in Czapek's solution agar produced colonies sulfur yellow, forming a close mycelial felt 200 to  $300\mu$  deep, with apparent development of perithecia or sclerotia with reverse and agar pale to deep orange; drops abundant pale yellow; conidiophores slowly developed, few and scattered, not sufficient to color the colony; upon wort agar colonies wrinkled, green color fairly well distributed, but yellow in sterile areas; conidiophores about  $4\mu$  in diameter, pitted; metulae 10 to  $12\mu$  long; sterigmata 8 to 10 by 2 to  $3\mu$ , with long sharp points; conidia fusiform to subglobose, about  $4\mu$  in long axis.

If the culture as received, correctly represents Biourge's idea of this species it probably belongs with the Ascosporic *P. luteum* series in which following Derx, any particular culture might be conidial or ascosporic.



372. *P. sacchari* Ray Rev. Gen. Bot. 9: pp. 294-300; Plate 16, fig. 23-27, 1897. See Sacc. Sylloge 22: 1276, no. 41.

Colonies upon sugar solution forming a thin felt of yellow mycelium in six days, with perithecia fairly abundant as more or less definite ascus-producing masses imbedded in the mycelium; vegetative hyphae about  $2\mu$  in diameter, sparingly septate, mycelial masses yellow to orange above and below; conidiophores erect about  $4\mu$  in diameter; penicillus consisting of two superposed verticils each of 4 to 5 metulae with their sterigmata and conidial chains, figured to show the symmetrically biverticillate characters clearly; sterigmata alternate at apex; conidia green 2 by  $1\mu$ ; asci six-spored, thin walled; ascospores 3 by  $2.5\mu$  thick walled.

Species found as yellow mycelium lining the channels made by borers in sugar cane from Martinique.

Ray's species has not since been reported. His figures fix it as symmetrically biverticillate; the pigment discussed, allies it with other members of the *P. luteum* series; the ascospores reported as six to the ascus and 3 by  $2.5\mu$  are smaller than those of related species described but it is entirely possible that it may be encountered again.

373. *P. spiculisporum* Lehman. Mycologia 12 (1920) 5, p. 268-274, pl. 19, figs. 1-37.

Colonies on potato or bean agar white or gray or with few and small greenish conidial areas, becoming cream, yellowish or pinkish with the ripening of the perithecia; surface growth consisting of floccose interlacing mostly creeping hyphae 2 to  $3.5\mu$  in diameter, bearing scattering conidiophores 10 to 50 usually less than  $20\mu$  long by 2 to  $2.5\mu$  in diameter, bearing a single verticil of few, 2 to 5, sterigmata 11 to  $16\mu$  long of uneven length in the verticil with very long acuminate points, occasionally borne singly or scattered in two's or three's, conidia 2.5 to  $4\mu$  by 1.8 to  $2.5\mu$ . Perithecia subglobose, 0.4 to 2 mm. in diameter, indehiscent, with wall consisting of woven hyphae, beginning to appear in about two weeks and ripening ascospores in 20 to 30 days, persistently white or becoming cream, pink and yellow shades in ripening. Asci elliptical, pyriform to globose 7.2 to 10.8 by 6.3 to  $7.7\mu$ , hyaline, 6-8 spored, with walls disappearing as the ascospores ripen. Ascospores ovate or elliptical, 2.5 to 4 by 1.8 to  $2.8\mu$  with walls minutely spiny (visible only with oil immersion and best after staining with carbol-fuchsin).

Culture: By Lehman; our no. 4391 is Lehman's own culture; it has

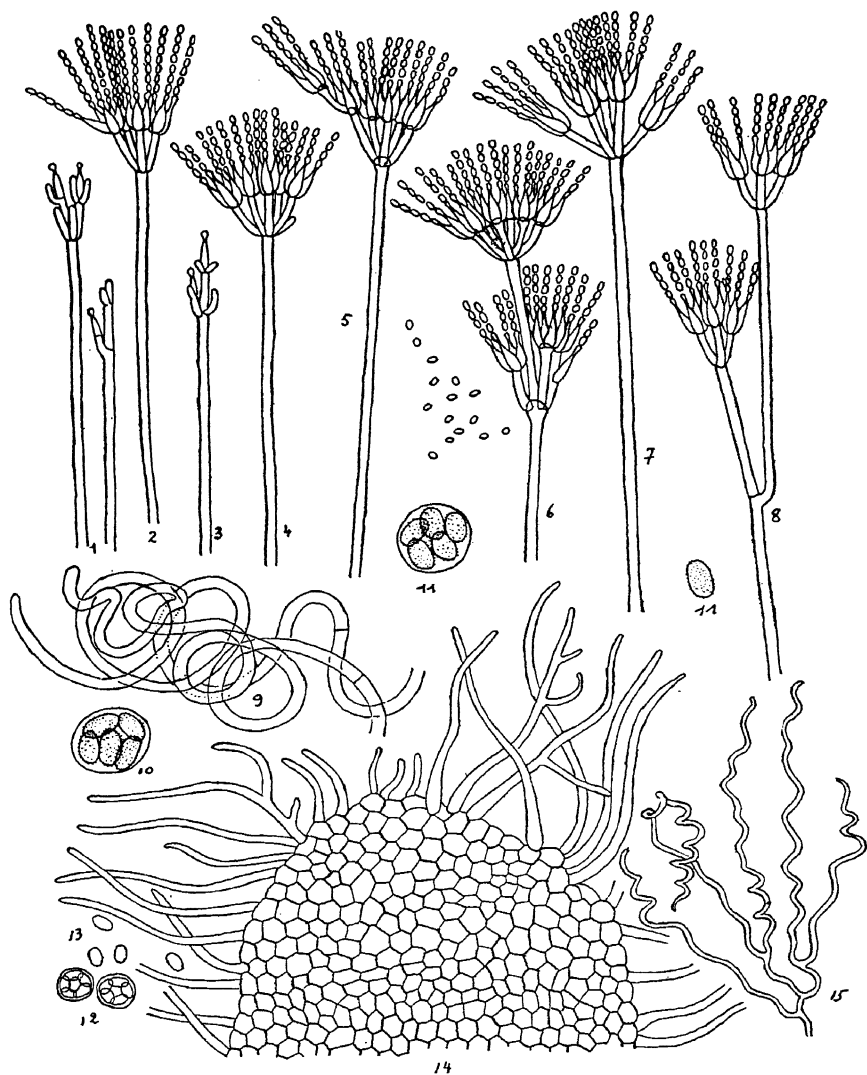


FIG. 71. Bainier and Sartory's figures of *P. petchii*: With detail of penicillus, ascus, and part of perithecial wall.

also been obtained independently in cultures from soil. In a culture of the type species developed in darkness, the perithecia were "canary" yellow when first seen but faded in sunlight within 2 hours to the color already associated with the species.

374. *P. aureum* Van Tieghem. Bul. Soc. Bot. France 24:157. 1877.  
Probably not *P. aureum* Corda.

Van Tieghem's organism produced conidia 3 by  $1.5\mu$ , with fruiting hyphae "golden" and ascospores yellow, oval, smooth, 5 by  $3\mu$ . While probably some member of this group, the species is not recognizable from the data given.

375. *P. petchii* Sartory and Bainier. Ann. Mycol. 11: pp. 272-277, Pl. XIV, fig. 1-12. 1913. See fig. 71.

Colonies upon licorice sticks wooly with crowded long conidiophores at first white then canary yellow becoming blue green (C.d.C. 367) in the conidial areas with margin remaining canary yellow as well as perithecial areas or masses; conidiophores crowded, up to 5 mm. long by  $5\mu$  in diameter; metulae 10 to  $12\mu$  long and 4 to 6 in the verticil; sterigmata 9 to  $10\mu$  long in verticils of 3 to 4; conidia elliptical or oval up to  $4\mu$  in long axis, swelling to double size in germination and producing 2 or 3 tubes; perithecia clustered, in beautifully yellow masses, 100 to  $200\mu$  in diameter, thin walled, with asci subglobose 12 to  $13\mu$  in diameter; ascospores, echinulate all over, about  $6\mu$  long, 6 in the ascus, suggesting the spores of the truffle.

Species found upon coagulated caoutchouc from South America. Optimum temperature  $26^{\circ}$  to  $28^{\circ}$ , maximum  $38^{\circ}$  to  $39^{\circ}\text{C}$ . The yellow coloring material was soluble in alcohol, glycerine, alcohol-ether, sulphuric ether but not in water.

A species resembling this has been received from Dr. Bottomley Pretoria, South Africa (4861D) and other forms not widely different occur occasionally as contaminants of other *Penicillium* cultures in this laboratory.

*Perithecia reported but ascospores not described.*

376. *P. gilvum* Sopp. Monogr., pp. 167-169, Taf. XI, fig. 130; Taf. XVIII, fig. 125; Taf. XXIII, fig. 21. 1912.

Species between *P. parasiticum* Sopp and *P. citrinum* Sopp with colonies floccose, dirty yellow developing scanty gray greenish yellow conidial

areas, and abundant reddish yellow globose, smooth; perithecia (or sclerotia ?-C. T.) enmeshed in loose hyphae; mycelium more or less yellow in most substrata; conidiophores moderately large figured as either typically biverticillate or with one distant, divergent branch producing a second penicillus, described as commonly *Aspergilloides*-like (Monoverticillate), but not so figured; metulae figured as short rather coarse, crowded and the verticils of sterigmata as producing parallel groups of conidial chains; conidia globose, yellow-brown,  $3.5\mu$ ; ascospores not described.

Species found in earth in west Norway. Colonies grew best at  $20^{\circ}$ , with minimum at  $3^{\circ}$  and maximum at  $30^{\circ}\text{C.}$ , and grew well on all media tested. Metallic odor production was noted in milk, but not on other media. Various substrata, milk, rice, were colored yellow. Conidia remained viable in the laboratory more than three years.

No one has ventured to identify this species.

377. *P. parasiticum* Sopp. Monogr., pp. 164-166, Taf. XII, fig. 127; Taf. XVIII, fig. 129; Taf. XXIII, fig. 19. 1912.

Colonies on meat-peptone-sugar-gelatine with mycelium intensively yellow, with clear yellow green conidial areas produced only when parasitic upon an insect larva and only perithecia upon sulphur yellow mycelium in cultures; conidiophores moderately large, figured as bearing 1 or 2 side branches at different levels, with penicillus consisting of a verticil of 6 to 12 metulae, sterigmata and diverging chains of conidia; sterigmata few in the verticil; conidia smooth, oval, 3 by  $4\mu$ , or occasionally globose; perithecia with a thin wall of dark green almost black cells, and central mass yellow were developed in large clusters and ripened spores only once in culture in which the ascospores were smooth, oval, yellow and larger than the conidia.

Species found upon an insect larva in soil; cultures grew best at  $20^{\circ}$ , with minimum at  $5^{\circ}$  and maximum at  $33^{\circ}\text{C.}$ , grew well in Sopp's gelatine, but not in his agar, grew very well on potato, but poorly or slowly in milk, wort, broth, rice and urine.

No one has reported this species since its publication.

378. *P. Kiliense* Weidemann. Centralb. f. Bakt., etc., 2 Abt. 19:680-683, fig. 1. 1907.

Colonies on sugar gelatine, and wort gelatine, strongly growing, green on the third day, with a gradual transition from sterile margin to conidial area, not a sharply marked white zone, on potato with marginal zone at

first white then yellow, becoming dark gray in age; characteristic in general as a flat close felted colony blue green in color, vegetative hyphae 2.4 to 3.5 $\mu$  in diameter; reverse colorless to yellowish; yellow color imparted to potato, rice and milk; gelatine not liquefied or very slowly liquefied in very acid media; conidiophores about 3 $\mu$  in diameter; metulae 10 to 12 by 3.5 $\mu$ ; sterigmata up to 10 $\mu$  long; with short tubes hence doubtfully related to the biverticillate series, conidia usual limits, 3.3 to 3.8 by 3 to 3.3 $\mu$ , clear green under the microscope, wall thin, clearly defined, in long chains, connectives visible, swelling in germination to 6 to 7 by 5 $\mu$ , and producing 1 tube; sclerotia yellow, smooth, up to 0.5 mm. in diameter, consisting of a broad yellow outer zone of pseudoparenchymatic hyphae with dark green special hyphae in center whose detailed structure was not further studied.

Milk showed dark blue green colonies with yellow mycelium on the margin and on the glass but digested milk remained uncolored.

Weidemann figured the penicillus as asymmetrical and gave details which scarcely belong with the biverticillate series but the presence of yellow sclerotia or developing perithecia is so characteristic of the group that any one finding this species again would probably automatically look for it in this group first. Its real relationship is problematical.

379. *P. citrinum* Sopp. Monogr., pp. 166-167; Taf. XII, fig. 128; Taf. XVIII, fig. 126; Taf. XXIII, fig. 21. 1912.

Colonies in meat-peptone-sugar-gelatine sulphur yellow with a greenish tinge, or in denser conidial areas blue green; hyphae fairly coarse, bright yellow; conidiophores comparatively coarse, enlarging slightly upward but not vesiculose at apices, with penicillus figured as 1 diverging branch then a typically biverticillate penicillus on the main stalk and branch; conidia elongated (langlich) about 4 $\mu$  long, angular? (eckig); perithecia (not described hence ? sclerotia) large yellow green, bean-shaped, once only seen.

Species parasitic upon *Polyporus vaporarius* and *Merulius lacrymans*. Cultures grew best at 20°, with minimum at 3° and maximum at 38°C., produced good colonies on Sopp's gelatine and agar, on broth, on potato.

No one except Sopp has reported this organism.

*P. virido-bruneum* Sopp. Monogr. pp. 200-201, Taf. XX, fig. 150; Taf. XXIII, fig. 42, 1912.

Colonies in meat-peptone-sugar-gelatine, at first white, quickly clear green to yellow green with the development of conidia which form a

thinly developed layer, mycelium thin, buckled, wrinkled; hyphae very coarse, septate; reverse clear white; odor in various substrata suggests formaldehyde; conidiophores coarse, jointed with walls granular toward the apex, resembling *Aspergillus* in vesicle-like enlargement toward the apex, with the production of a dense verticil of obpyriform or clavate metulae with crowded verticils of sterigmata on conidial chains; sterigmata sharply pointed; conidia oval  $4\mu$  by  $2.5$  to  $3\mu$ .

Perithecia formed in cultures on bread, clear gray brown, consisting of irregular, thin walled parenchyma-like cells, but Sopp failed to give the essential data as to ascospore formation.

Species found in soil. Cultures grow best at  $20^{\circ}$ , with minimum at  $5^{\circ}$  and maximum at  $37^{\circ}\text{C.}$ , and grew well on milk which was colored yellow, in urine, in broth, in wort, on potato, on rice which was colored yellowish green, then red-brown and upon bread.

No one has recognized this species since it was published.

381. *P. niveo-rubrum* Sopp. Monogr., pp. 190–192; Taf. XI, fig. 140; Taf. XVIII, figs. 139, 141; Taf. XX, fig. 142. 1912.

Colonies on meat-peptone-sugar-gelatine showing two forms at different seasons, (1) in the autumn a chalk white mycelium spreading broadly and smoothly over the substratum in which reddish perithecia appear and finally develop into a crust composed of crowded perithecia first salmon color then blood red which ripen slowly; in the other form (2) in spring, summer and winter, a thick tough mycelium sordid grayish yellow below, much wrinkled, becoming green to yellow green with ripening conidia, and showing moderately long and coarse conidiophores, clavate at the apex and bearing single or superposed verticils of metulae or sterigmata, variously arranged and variously branching but maintaining the general appearance of the biverticillate group. These forms are found mixed in the same colonies in intermediate seasons; conidia  $3.5$  to  $4\mu$ .

Species from the earth in east and west Norway. Cultures grew best at  $20^{\circ}\text{C.}$ , with minimum at  $3^{\circ}$  and maximum at  $33^{\circ}\text{C.}$ , and grew well upon the common media of Sopp. No definite odor was produced, and no coloring matter. Conidia remained viable more than 3 years.

No one ventures an identification of this species.

*Section 2. Coremigena.*

385: *P. Duclauxi* Delacroix. Bulletin de la Société Mycologique de France, Tome VIII, 1891, p. 107, Pl. VII. See Thom, U. S. Dept. Agr. Bur. Amer. Ind. Bul. 118:42, fig. 9, 1900. See fig. 73.

Colonies grown upon gelatin and potato or bean agar clear dark green to olive when old, consisting of short crowded conidiophores arising for the most part singly from the substratum (strict), but sometimes producing short coremia; long coremia are produced abundantly upon orange, milk, potato, and all media rich in cane sugar; conidiophores very short, 10 to 50 $\mu$ , either arising directly from the substratum or borne upon the upper third of the coremia, 1 to 2 septate, bearing a simple conidial fructification or a terminal fructification and a divergent

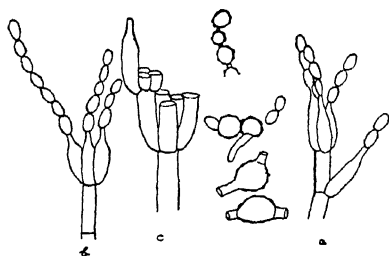


FIG. 72. *P. duclauxi* Delacroix: Detail of metulae, sterigmata and conidia from Thom, 1910.

lateral branch with a whorl of sterigmata; penicillus often 100 to 160 $\mu$  in length consisting of a few sterigmata 10 to 12 $\mu$  in length in a simple terminal whorl or less commonly in secondary whorls; conidia elliptical fusiform 3.6 to 4 by 2 to 2.5 $\mu$ , clear homogeneous green, smooth when young, but rugulose when ripe; colonies liquefy sugar-gelatin in Petri dish culture slowly from twelfth to twenty-fifth day and change red litmus to blue in seven days; produces a coloring agent in sugar media which is wine red in alkaline media and yellow (bile-yellow) in acid media (acts as an indicator with neutral point very near that of phenolphthalein).

This fungus is characterized by its enormous development of coremia upon milk, orange, apple, and media containing cane sugar, while producing only very short conidiophores in bean or potato agar and gelatin free from sugar.

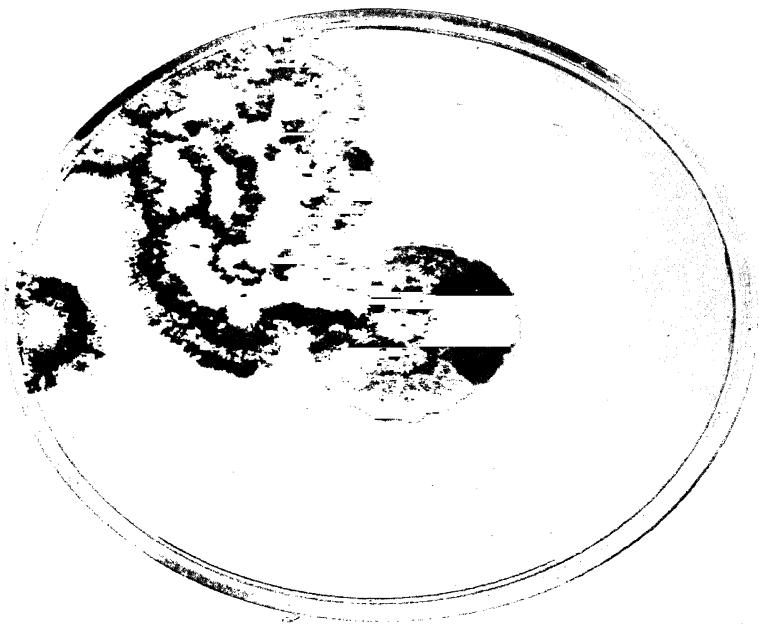


FIG. 73. *P. duclauxi*: Photograph of colony showing ridged masses of white coremia in the early stage of fruiting.





Culture no. 20, received from the author, Georges Delacroix, Paris.

The species has since been encountered many times hence is believed to be present in many different parts of the world.

Biourge in Monogr. La Cellule 33: fasc. 1, p. 248-250; Col. Pl. XI, Cart. 351; Pl. XVII, fig. 101; 1923, reported in a reference on p. 98, that Dierckx obtained *P. duclauxi* from Delacroix. The culture probably passed to Biourge directly. Biourge's no. 351 (4733.53) was received in September, 1927, and is evidently correctly named although he adds little to previous discussions. In cultures, four months in the laboratory and showing abundant coremia the stalks were yellow in age thus suggesting the possibility that this species which is now known to be widely distributed may have been the basis of Corda's *Coremium citrinum* or of *P. bicolor* Fries.

*P. duclauxi* was isolated by us from a peculiar appearing culture of *P. expansum* received from C. C. Barnum at Berkeley, California. In the original mixture the usual characters of *P. duclauxi* were almost completely suppressed but its presence was reflected in the colors produced, and in the structure of the coremia. This lead to careful selection of conidia which developed normally while the culture of *P. expansum* lost its peculiarities.

*P. bicolor* Fries. Sys. Myc. 3, p. 408. 1829.

Significant data freely translated from the Latin: Sterile hyphae effused, yellow; fertile hyphae aggregated into fascicles, penicillate at the apex; spores glaucescent; separated from the other species of the genus on account of the colored vegetative hyphae; fertile hyphae at first forming a tubercle then elongating to produce a stipe with a subglobose head.

No warrant is found for applying this name to a particular form, but the contrast noted by Fries between the sterile hyphae as yellow and the fertile hyphae which carried the green color is fairly characteristic of this whole section of the biverticillate group. An occasional culture is encountered which might readily have formed the basis of Fries' description.

Organisms identified under this name have been reported by Oudemans (Nederl. Kruidk. Arch. Ser. 2: 1123, 1904), from earth at Utrecht, who gives conidia globose  $2.3\mu$ ; by Goddard, Bot. Gaz. 56: 268, fig. 6B, 1913, but without evidence of valid identification.

Fries' species probably belonged about where we have placed it among the coremiform biverticillate series. Among the synonyms he cited

*Monilia penicillus* Persoon Obs. Myc. 2, p. 35 Table 4, fig. 9; *Coremium bicolor* Liljebl, *Coremium glaucum* and *citrinum* Pers. Myc. Eur. 1, p. 42-43.

387. *P. Zukalii* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 239-240; Col. Pl. VII, Cart. 198; Pl. XI, fig. 61. 1923.

Colonies on wort gelatine obscurely zonate, blue green, then green, finally gray green; coremia common with stalk variously white, orange, red to dark brown; on other media stalks may be very short or almost absent, as on bean agar; reverse yellow to red, brown and finally dark brown, with margin carmine or orange; odor none, or feebly ammoniacal; conidiophores about  $4\mu$  in diameter, with walls smooth; penicillus figured as simple biverticillate forms up to  $20\mu$  long, or more complex from the development of branches from lower nodes or the proliferation of metulae, hence doubling the length to 35 to  $40\mu$ ; branches occasionally produced, 12 to 20 by 3 to  $3.5\mu$ ; metulae 14 to 22 by 3 to  $3.5\mu$  in twos, threes, or fours; sterigmata 11 to 18 by 2.5 to  $4\mu$ , in twos, threes, or fours; conidia at first round, presently elliptical 3 to 6 by 3 to  $4\mu$ , very slightly echinulate; perithecia not found.

Biourge's no. 198 (our no. 4733.128) was recorded as related to *P. rugulosum* Thom, also to *P. atramentosum* (? C. T.) and to be one of Direckx's unpublished "Diversiramosa."

We do not find any *Penicillium* in which the conidia change from globose to elliptical; an apparent change of this kind may be suggested by the very different rate at which conidia change shape under successive conditions of culture. As a result some older conidia may show pronounced ellipticity and perhaps be arrested at that stage and fail entirely to become globose, while younger conidia may pass the elliptical stage more quickly and reach a subglobose or globose form.

An occasional culture shows characters suggesting this species (no. 4360); in others coremium formation is reduced to aggregations of conidiophores without the clear development of coremia. These species are assigned to *P. sulfureum*.

*Section 3. Luteo-virida.* Colonies commonly showing green conidial areas on mycelium which shows more or less yellowing (from yellow granules); reverse, agar or both commonly showing yellow, orange, red or a succession of shades from yellow to red; ascospores and sclerotia not formed; erect coremia usually wanting.

*Sub-section 3a. Funiculosa:* Prostrate or ascending (scarcely erect) ropes or funiculose bundles and anastomosing networks of aerial hyphae regularly present and characterizing the colony (fig. 74).

*Sub-section 3b. Luteo purpurogena.* Conidial areas green usually interspersed at some stage of growth with yellow or reddish (in age) hyphae especially in the marginal areas; ropiness reduced inconspicuous or absent; reverse usually in yellow or orange to red colors (rarely uncolored).

*The Luteo-viride-pinophilum series.* Many variant strains show the funiculose colony with the brilliant yellows and rich greens described for *P. luteo-viride* and *P. pinophilum*.



FIG. 74. The funiculose colony with biverticillate penicilli: From no. 4658.174.3, provisionally *P. luteo-viride* Biourge (no. 390).

390. *P. luteo-viride* Biourge. Monogr. La Cellule 33: fasc. 1, pp. 242-243; Col. Pl. VII, Cart. 64; Pl. XI, fig. 62. 1923.

Colonies on wort gelatine restricted in growth, almost coriaceous, from blue green to dark olive green, with narrow margin yellow, inside an outermost white zone (submerged); coremia none; reverse at first orange, then variegated or polychromatic; gelatine liquefied and yellow; odor none; conidiophore about  $3\mu$  in diameter, with walls smooth, figured as short branches from trailing hyphae; penicillus about  $20\mu$  long, figured as symmetrically biverticillate or as consisting of short monoverticillate branches from trailing hyphae; metulae 7 to 12 by 2 to  $3\mu$ , commonly in fours, figured in one case as granular; sterigmata 9 to  $11\mu$  long in verticils of 3 to 5; conidia elliptical  $2.5$  to  $4\frac{1}{2}$  by 2 to  $3.2\mu$ .

Biourge's type no. 64 was not received. Biourge's no. 98 (our no. 4733.84) was received in September, 1927, as *P. luteo-viride* but did not fit the above description. Subsequent purification showed this culture to contain *P. citreo-viride* Biourge and an unidentified organism which was not a *Penicillium*.

An organism suggestive of *P. luteo-viride* Biourge no. 4658.174/3 was received from Putterill at Cape Town in 1923. Our notes follow: Colonies dense dark green in the central area, zonate and with radiating wrinkles; margin broad passing from white at the very edge through pale yellow to green; aerial growth bristly fasciculate or ropy bundles anastomosing to form networks; reverse in yellow or sometimes greenish shades becoming brown in the older areas; drops deep dark green; conidiophores up to  $100\mu$  long mostly very short branching from trailing hyphae and ropes of hyphae; penicilli variously but more or less typically biverticillate; conidia 3 to 3.5 or even  $4\mu$  by  $2.5\mu$ , rough.

This strain was maintained for a time in culture but eventually lost.

391. *P. pinophilum* Hedgecock. In Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, p. 37-8, fig. 6. 1910. See our figs. 75, 76.

Synonym: *P. aureum* Corda. Emended Hedgecock, Mo. Bot. Gard. Rept. 17, pp. 105-107, pl. ii, figs. 1-3.

Not *P. aureum* Corda. Prachtflora, p. 38, t. XVIII.

Colonies on potato or bean agar and milk sugar gelatin, from green on agar through shades of yellow-green to bright yellow and orange on media containing starch and cane sugar. Superficial hyphae studded with yellow granules upon acidified media. Reverse of colony and substratum (upon these media) colored deep rich red. Surface growth partly of simple conidiophores, partly aerial hyphae, and ropes of hyphae (which rarely become vertical coremia) bearing conidiophores as lateral branches. Conidiophores 100 to  $200\mu$ . Penicilli up to  $120\mu$  in length, consisting of single verticils of metulae 10 to 16 by 2 to  $2.5\mu$ , bearing whorls of sterigmata cells 13 to 15 by 2 to  $2.5\mu$  tapering, acuminate, bearing conidial chains which are parallel but do not form a column. Conidia elliptical, 3 to 3.6 by  $2\mu$ , smooth, pale green or yellowish green. Colonies liquefy gelatin, but slowly and incompletely, and give a neutral or acid reaction upon all litmus media. Under different conditions of culture and acidity the discoloration of the medium varies from yellow to orange and deep red. Produces discolorations upon commercial timbers.

Habitat: Pine wood, which is strongly colored by it.

Culture received from the author, G. G. Hedgcock, of the Forest Pathological Laboratory, Bureau of Plant Industry, United States Department of Agriculture.

*P. pinophilum* was reported by Hedgcock as a cause of discoloration in pine wood. We have isolated organisms indistinguishable from this species from various sources, hence conclude that this represents a fairly common variant in the biverticillate and funiculose series.

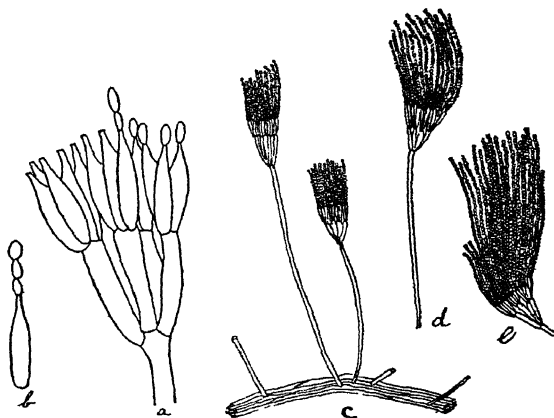


FIG. 75. *P. pinophilum* Hedgcock: Detail of penicillus *a* and one sterigma *b*; *c*, *d*, *e*, sketches of penicilli with a rope of hyphae.



FIG. 76. *P. pinophilum*: Diagrammatic radial section of margin of colony (magnified 25 times).

Among the strains listed we have no. 4202.16T from Professor Thaxter; no. 4695.11 from Miss Derick at Montreal; one from cornstalks at Alexandria, Va.; no. 4861 from South Africa; no. 4303.69 from Louisiana; no. 4971 from a paste pot at Washington.

*P. humicola* Oudemans as interpreted by Gilman and Abbott belongs in this series but can not be identified as Oudemans's species from the description.

*P. funiculosum* series. Numerous cultures have shown the general

characters of *P. funiculosum* with detailed differences in mass of growth or intensity of color reactions: Nos. 392, 393, 394, 395.

392. *P. minio-luteum* Dierckx. Soc. Scientifique Bruxelles 25: p. 87. 1901. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 237-239; Col. Pl. VII, Cart. 60; Pl. XII, fig. 67. 1923.

Probable synonym: See Biourge: *P. variabile* Sopp, (No. 1 C. T.)

Colonies on wort gelatine, rather tough, almost velvety, green, dark green, in age dark brown, coremia tardily produced red at base; reverse yellow then orange shades, to red (miniata), drops red, odor none; conidiophores about  $3\mu$  in diameter, with all walls smooth except conidial wall, slender and graceful fruiting mass; penicillus figured as strictly biverticillate and about  $25\mu$  long, or 40 to  $65\mu$  by the development of an accessory complete or partial verticil of metulae lower down; metulae 9 to 14 by 2 to  $2.8\mu$ , in verticils of 4 to  $8\mu$ ; sterigmata 10.5 to 14 by 2 to  $2.5\mu$ , commonly in threes, occasionally more numerous; conidia 3 to 4 by 2 to  $3\mu$  tardily echinulate, commonly fusiform.

Biourge no. 60 (our no. 4733.89) when grown upon Czapek's solution agar produced colonies deep green, with funiculose areas, about  $600\mu$  deep, with more or less yellow aerial hyphae throughout, especially when young and persisting during the growing period at the margin; producing yellow to orange drops; reverse yellow to deep red, with agar paler red than the mycelium.

These minor differences are incident to the change of substratum.

393. *P. funiculosum* Thom. U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118: 69. fig. 27. 1910. See fig. 77.

Cultivated in gelatin or potato or bean agar, deep green, broadly spreading, surface closely floccose with procumbent hyphae, tufts and ropes of hyphae bearing lateral conidiophores; reverse becoming red, purple, or very dark purple, almost black, with the whole mass of medium colored; conidiophores short, 20 to 80 or  $100\mu$ , mostly perpendicular branches from trailing hyphae, sometimes arising separately from the substratum; penicilli up to 125 or  $160\mu$  in length, with 1 or 2 alternate appressed branches bearing verticillate branchlets and dense verticils of parallel sterigmata 10 to 14 by 2 to  $3\mu$ ; conidia at first cylindrical, then elliptical or fusiform, 3 to 4 by 2 to  $3\mu$ , green, in chains which break up completely in fluid mounts; colonies not liquefying gelatin in 2 weeks, with acid reaction to litmus.

Type Thom no. 42 found in accidental culture, Storrs, Conn., 1905;

also received from Dr. E. A. Bessey, Miami, Fla., 1908. Cultures scarcely distinguishable from no. 42, have been received from a number of different sources, for example, no. 4270D from Chung in China; no. 4861 two cultures from Pretoria, South Africa, no. 4341.4L6 from Miss Walker, Lincoln, Nebraska.

394. *P. fastigiatum* nomen nudum. G. F. Atkinson, no. 14910 in Flora Cayuga Lake Basin, New York; Cornell University.

Label reads "Hyphae erect 80 to 200 by 5 to 6 septate, spore bearing fastigiately branched branches extending close against the main hyphae and branches also crowded. Branches usually monopodial, tips sometimes opposite. Terminal spore bearing branch narrowly terete, 12 to

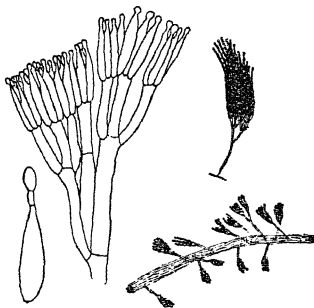


FIG. 77. *P. funiculosum* Thom: Detail of penicillus with verticils of long closely packed sterigmata and ropes of hyphae producing conidiophores.

20 by 1 to 1.5 $\mu$ . Spores in chains, oval to short oblong. Smooth, 1 to 3 by 1 to 2 $\mu$ . G. F. A."

Thom and Church no. 4140. This herbarium specimen when examined in 1916 was not viable, but microscopic examination showed spores, smooth, transparent, globose, more than 1.8 $\mu$  in diameter, and was tentatively placed from the description on the label with *P. funiculosum*.

Habitat: On wood.

Not cultured by Atkinson.

395. *P. africanum* Doebelt. Ann. Mykol. 7: 315-338. 1909.

Doebelt gave: Colonies in sugar, mineral salt agar at first gray white then green (producing conidia within two days), becoming dark green,



with an outer zone yellow surrounded by a white mycelial margin, finally gray brown; reverse and agar red; conidia oval 2.8 by 2.4 $\mu$ .

Species obtained from sugar cane in Africa.

Doebelt although giving the biochemical reactions of his species failed to describe the structures further. His culture (our no. 104) was lost, from our collection, but was again received from Pribram (no. 4777.5) in 1923, it shows his species to belong to the biverticillate series in the section in which the surface growth consists of anastomosing ropes of hyphae bearing conidiophores as short branches. It is closely related to Thom's *P. funiculosum*.

Biochemical reactions as given by Doebelt include: red color produced in mineral salt medium with the following sugars, arabinose, dextrose, levulose, galactose, glucose, raffinose; growth not restricted by 20 per cent cane sugar but much restricted at 40 per cent; growth was weak and colorless in alkaline media.

396. *P. islandicum* Sopp. Monogr., pp. 161-164, Taf. XVII, fig. 122; Taf. XXIII, fig. 25-26. 1912.

Colonies characterized by their three-colored appearance, yellowish green conidial area in the center surrounded by a yellow zone bordered reddish; mycelium above yellow, close woven, heavy stereum-like on wort media; conidiophores figured as smooth, slender, 60 to 80 $\mu$  long, occasionally branched far from the apex, with penicillus consisting of 1 verticil or 2 superposed verticils of metulae and sterigmata with parallel conidial chains (not divergent, not in columns; conidia oval, 1.5 to 2 $\mu$  by 3 to 3.5 $\mu$ ; perithecia not found.

Species found on the Island of Skyr and a variety upon Stereum in Norway. Cultures grew best at 20° to 25°C., continued to grow at 8°C. and at 38°C. The species was selective in its response to media growing well upon Sopp's gelatine and agar, acid broth, wort, potato, rice and bread, but less readily upon milk and neutral broth; conidia remained viable more than three years.

Two cultures, no. 4658.35.1 and no. 4658.35.144, from Putterill in South Africa reproduce this description closely enough to justify identification although the original was not distributed by Sopp. Other strains of the same series have since been found.

Putterill's cultures may be described (no. 4658.35.1 and no. 144.2): Colonies dense, tough, bristly, at first, showing zones of orange and lighter color, with margin paler, later developing a green conidial zone near the margin and progressively overgrown with red orange to red

hyphae in the central areas, reverse at first orange to sordid yellow orange shades, later becoming rich red shades, with the agar partly colored in the same or paler shades; conidiophores usually restricted to marginal zones, arising from submerged hyphae, producing biverticillate penicilli and conidia elliptical with long axis  $4\mu$ .

*P. herquei* series. The strains of this series studied amply justify the belief that Sopp's name *P. elegans* was used for one of them; includes nos. 397, 398, 399, and 400.

397. *P. herquei* Bainier and Sartory. Bul. Soc. Mycol. France 28, fasc. 2, pp. 121-126, Pl. VII. See also Sartory and Bainier, Compt. Rend. Soc. Biol. Paris 71: pp. 229-30, 1911 (our fig. 78).

Characterization emended: Colonies on Czapek's solution agar with mycelium more or less floccose with some trailing hyphae and ropes of hyphae, white then golden yellow (hyphae covered first with drops then crusted with yellow granules) becoming yellow-green with the development of conidial areas; reverse and agar yellow (C.d.C. 206), with color extending rapidly beyond the limits of the colony; vegetative hyphae closely felted 1 to  $3\mu$  in diameter; odor none; conidiophores up to 500 even to  $1000\mu$  long by 3 to  $3.5\mu$ , septate, unbranched, but occasionally anastomosing with adjacent conidiophores, with walls heavily encrusted with yellow granules (? pitted), producing at the apex a crowded, compact, verticil of metulae; metulae 8 to  $10\mu$  long; sterigmata about 7 to  $9\mu$  long, tapering rather abruptly to a narrow conidia-bearing tube; conidia subfusiform, delicately roughened, from 3 by 2 to 3.5 to  $4\mu$  by  $2.5\mu$ , occasionally doubled in size, very slightly colored, in loosely divergent and tangled chains; perithecia not found.

Species found among leaves of *Agauria pyrifolia* sent by Col. Herque to l'École Supérieure de Pharmacie, at Paris. The diagnosis presented combines the information given by the describers with the data from our own cultures since we have studied at least five strains; no. 4640.447 which was Bainier's Type culture, no. 4136H38 from New Jersey soil, no. 4445 from an insect, no. 5001.15b from soil at Evanston, Illinois, and no. 4303.66 from Louisiana, no. 4897.47 from Naganishi Dairen Manchuria. Cultures grew well in all media with temperature range given by the describers at  $15^{\circ}$  to  $38^{\circ}$  with optimum  $26^{\circ}$  to  $28^{\circ}\text{C}$ . Gelatine was not liquefied. Where peptone was used (see Sartory and Bainier, loc. cit.), the medium became green rather than yellow. The coloring substance is soluble in all the solvents of fatty substance.

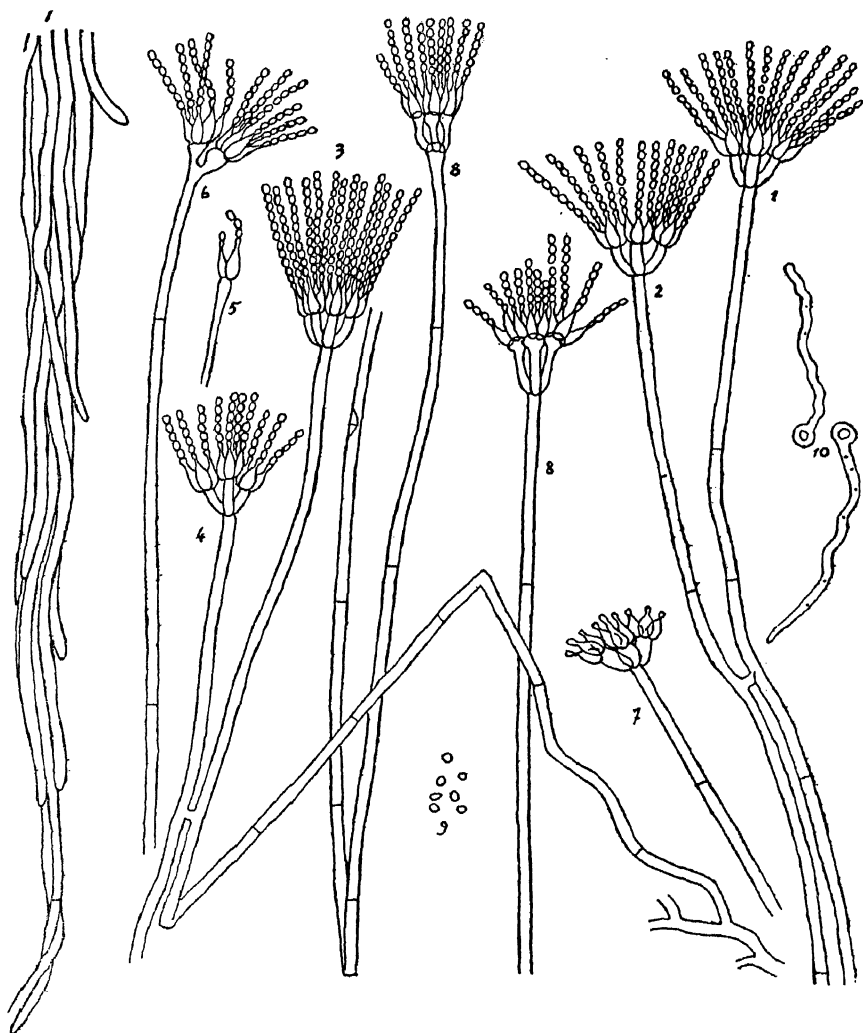


FIG. 78. Bainier and Sartory's plate of *P. herquei*: The futility of detail figures in this group is fairly clear when we compare the figures of *P. petchii* and *P. herquei*.

Biourge received no. 4640.447 from us (Biourge 1925) but does not seem to have understood its significance as the type of *P. herquei*.

398. *P. aureum* Corda. Prachtflora, pp. 37-38, Taf. XVIII, fig. 1-3.  
1839. Not Biourge, Monog. 111-114, 1923, for which see no. 270.

Description compiled from Corda's Latin diagnosis, his German comments and his figure: tufts small, in groups golden yellow to parrot-green then olivaceous; conidiophore erect, long, graceful, septate, described as olive green, figured as producing branches opposite (in pairs) and cruciate at the upper 2 to 3 nodes, but probably representing superposed verticils of metulae; conidia small, figured as elliptical and in long diverging chains, described as first golden yellow then parrot-green.

Species found in cracks and clefts and upon "worm dust" in a rotten pine water spout in a moist place in a garden in Prague, 1838.

The description given, the substratum, and the figure place this as a member of the biverticillate section and not far from *P. herquei* B. and S. in which the rich development of yellow granules on the conidiophore and its branches would easily account for the golden colors reported by Corda. Identification with Biourge's no. 144 is rejected because that species can not be made to satisfy Corda's description except as to the "parrot-green" color. Biourge did not have *P. herquei* and its allies in culture hence had not seen the fitness of placing *P. aureum* with that group of species.

399. *P. lemoni* Sopp. Monogr. pp. 194-196; Taf. XX, fig. 152, Taf. XX, fig. 152; Taf. XXIII, fig. 39. 1912. Compare *P. herquei* Bainier as a possible synonym.

Colonies on meat-peptone-sugar-agar, with strongly growing, thin, whitish, wrinkled mycelium, with a fine surface network of hyphae, dull green, to yellow green; reverse somewhat yellowish to reddish becoming brownish in old cultures; gelatine liquefied and finally brownish; in young cultures producing a wonderful rose oil odor (ethereal oil odor); conidiophores coarse, few septate, rough, granular, enlarged upward, producing a verticil of metulae or branches, which are partly clavate, long and bear verticils of metulae, part (in Sopp's figure) bear verticils of sterigmata, as described manifestly biverticillate in aspect although the figures are doubtfully interpretable; sterigmata irregular in size; conidia 2 to  $3\mu$  in very long chains: perithecia (sclerotia) produced upon bread, rice, potato, gelatine, and wort cultures, large, consisting of

thick walled parenchyma cells, white with dark "vein" and points, and held together in clumps by reddish brown hyphae.

Species found on an orange peel. Cultures grew best at 20°C., with a minimum at 5° and maximum at 37°C., and grew well upon all of Sopp's test media.

Conidia remained viable more than three years. Sopp's type has not been seen.

C. J. Humphrey's culture no. 12719.7 from wood at Madison, Wisconsin, may be included in *P. herquei* in a broad interpretation of that species or perhaps furnish the bond between Sopp's description of *P. lemoni* and that species. The colonies upon Czapek's solution agar were closer textured, with shorter conidiophores in the central area, long ones only at the margin; the agar was yellowed; the reverse of the mycelium a deep delft blue.

The wonderful rose oil odor noted by Sopp might apply to the odor of this and some other cultures which repeat the morphology of *P. herquei*. Bainier did not report odor for his species but our strain from the Bainier collection has pronounced odor.

No perithecia or sclerotia have been found in our cultures but the coarse, rough stipe, the general type of branching shown in Sopp's figure, the odor as described and the broad distribution of the *P. herquei* series of cultures which we have seen, lead us to put *P. lemoni* with *P. herquei*.

400. *P. elegans* Sopp. Monogr., pp. 144-145, Taf. XVI, fig. 112; Taf. XXII, fig. 13. 1912.

Colonies broadly zonate, deeply blue-gray, becoming progressively more yellowish green, with a heavy white marginal zone; mycelium white in reverse, in age greenish yellow; hyphae coarse; odor strong, from petroleum-like to ether-like; conidiophores very long, uniform in diameter, coarse, septate, somewhat rough, branched, figured as producing a long partly divergent branch at some distance below the penicillus which bears a separate penicillus; penicillus figured as the main stalk with 1 branch then verticils of metulae; sterigmata very numerous, short, cylindrical; conidia 3.5 to 4 $\mu$  yellowish green; perithecia not found.

Species found upon wood in cellar-earth, with optimum temperature at 20°, minimum at 5° and maximum at 33°C. Colonies grew well on gelatine and agar, milk, urine, broth, wort, potato and rice. Conidia remained viable more than four years in the laboratory.

401. *P. olsoni* Bainier and Sartory. Ann. Mycol. 10: 398-9; Pl. VI, figs. 1-8. 1912.

Colonies on banana forming tufts bluish (compare C.d.C. 378), becoming grayish blue green (C.d.C. 372, 373) in age sordid yellow green; conidiophores erect, rigid, up to  $8.4\mu$  in diameter and comparatively very long, branches wanting or produced occasionally in age far down on the stalk and bearing a secondary penicillus; penicillus 2 to 3 times verticillate, usually symmetrically, occasionally with a superposed verticil on the main stalk prolonged; branches in the primary verticil (metulae?) up to 12 or more in number  $8.4$  to  $11.2$  by  $3.2$  to  $5.6\mu$ , bearing either a secondary verticil of shorter and smaller metulae or sterigmata 4 to 6 in the verticil  $8.4$  to  $11.2$  in length with long tapering points bearing the conidia; conidia ovoid, averaging  $3.2$  by  $2.8\mu$ ; sclerotia and perithecia not reported.

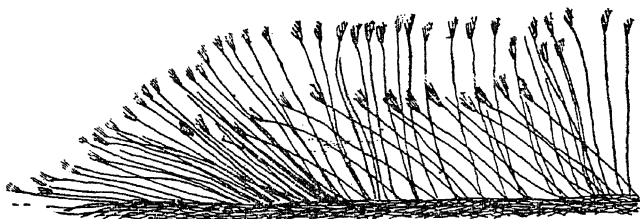


FIG. 79. *P. olsoni* Bainier and Sartory: Diagrammatic radial section of margin of colony (magnified 25 times); the crowded area of long conidiophores is characteristic here.

Species found upon the skin of a banana; cultures grew best at  $24^{\circ}$  to  $25^{\circ}\text{C.}$ , but grew up to  $35^{\circ}\text{C.}$

The culture under this name in the Bainier collection (no. 4640.450) was a member of the series *P. rugulosum* hence lacks the conspicuous morphological features described for this form. Culture no. 4725.1021S received from C. G. Hansford in Jamaica produces conidial fruits with very nearly the characters of *P. olsoni*. Our notes follow (fig. 79): Colonies in Czapek's solution agar spreading broadly in its conidial stage, bluish green, loosely velvety, forming a mass of loosely standing stalks up to 2 mm. deep, with rather thin margin, later with the development of an overgrowth of ropes and tufts and masses of hyphae with sporadically at least the development of sclerotia or perithecia, black, more or less submerged, brittle but so far as studied producing no asci; reverse pale yellow to red or reddish in areas; conidiophores long, coarse,

500 to 1000 or even 2000 $\mu$  by 8 $\mu$ , unbranched, colorless, smooth; penicillus biverticillate, or in some triverticillate, consisting of a crowded verticil of diverging metulae about 10 to 12 in length, and much smaller in diameter than the stalk, bearing the sterigmata or occasionally with secondary verticils of shorter cells 8 to 10 $\mu$  long, bearing the sterigmata; sterigmata up to 10 $\mu$  long, more or less long pointed; conidia elliptical to almost globose 3 to 3.5 $\mu$  in long axis smooth or with faint traces of granulation or spinulosity in the walls.

*Sub-section 3b.* In contrast to the series of non-ascosporic species in which funiculose hyphae constitute the dominant character of the cultures, there are three groups of species and strains in which these funiculose hyphae, while often present, are reduced or inconspicuous:

- (1) Colonies with conidial masses dark green commonly mixed with sterile hyphae encrusted with yellow granules and with reverse orange to red orange; conidia commonly rough or rugulose.....*P. rugulosum* and allies.
- (2) Colonies with reverse quickly deep purple red.....*P. purpurogenum* and allies.
- (3) Colonies with more or less conspicuous green conidial areas with sterile hyphae colorless or yellow (luteus) and reverse mostly in yellow color only tardily going over to orange or occasionally red.....*P. luteum* series (non ascosporic).

*P. rugulosum* series. A cosmopolitan group of strains with rough conidia and brilliant orange tints and shades in reverse: including nos. 405, 406, 407, and 408.

405. *Penicillium rugulosum* Thom. U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 60-61, fig. 21. 1910. (figs. 80, 81.)

Cultivated in Czapek's solution agar, yellowish green, then green, at length dark green; surface growth of densely crowded conidiophores with few aerial hyphae and even ropes of hyphae interspersed at their bases; reverse of colonies yellow to orange in spots, especially upon potato or upon sugar media; substratum not or slightly yellowed; conidiophores 100 to 200 by 2.5 to 3 $\mu$ , arising separately or branching from aerial hyphae just above the substratum; conidial fructifications 100 to 150 $\mu$  in length, consisting of appressed, verticillate branches 10 to 15 by 2.5 $\mu$ , bearing verticils of sterigmata, of metulae, or of sterigmata and metulae together; sterigmata 9 to 12 by 2 $\mu$ , acuminate, bearing long divergent chains of conidia; conidia 3.4 to 3.8 by 2.5 to 3 $\mu$ , ellipti-

cal, green, more or less apiculate at one end, verruculose when ripe, swelling to  $5\mu$  and germinating by one or two tubes; colonies do not or only partially liquefy gelatin.

Common in cultures, Storrs, Conn. Characterized by its verrucose spores and the brilliant color of the mycelium viewed from below.

The species is discussed by Biourge (Monogr. La Cellule 33, fasc. 1, pp. 246-248; Col. Pl. VII, Cart. 184; Pl. XII, fig. 68, 1923; his culture no. 184 was received in Sept. 1927 and showed some divergence in habit and color production from Thom's no. 46. Biourge notes in addition to Thom's description more or less septation in the metulae, the presence of occasional very large (giant) conidia.

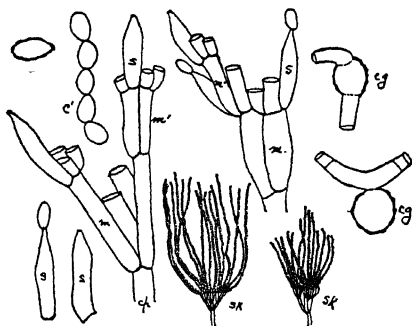


FIG. 80

FIG. 80. *P. rugulosum* Thom (figures of 1910): *sk*, Detail, sketches; *cg*, germinating conidia; *m*, metulae; *m'*, secondary metulae in verticils otherwise sterigmata *s*.



FIG. 81

FIG. 81. *P. rugulosum*: Diagrammatic radial section

Strains with the morphology and cultural reactions of *P. rugulosum* have been obtained from many sources. The species appears to be abundant in soil and soil contaminated products and to be very widely distributed.

As parasites of other species of molds, members of this series have been many times found in our studies of the Aspergilli. In the factory use of *Aspergillus niger* for fermenting sugar solutions Currie (personal communication) reports that a colony of this species will produce circular areas of infection in the floating blanket of *Aspergillus* mycelium which appear to rot and drop away when the mycelium is disturbed. In culture the *Aspergillus* mycelium is penetrated by dense mats of *Penicillium* hyphae which encircle the stalks and fruit as densely radiat-



ing conidiophores often completely obliterating the black heads of the *Aspergillus* with the gray green fruiting branches of the *Penicillium*.

405a. *P. verruculosum* Peyronel in I germi atmosferici dei funghi con micelio, page 22, Padova. 1913.

Colonies discoid, dense, at first yellow, then yellow green, finally dark green; sterile hyphae creeping or decumbent, septate, at first hyaline, soon yellow, at length orange; conidiophores erect or ascending, septate, yellow, 80 to 100 by 3 to 3.5 $\mu$ , with apex verticillately branched; branches (metulae?) 2 to many, 7 to 10 by 2.5 to 3 $\mu$ , bearing verticils of 3 to many sterigmata, 8 to 10 by 2 to 3 $\mu$ , with chains of conidia forming a column; conidia globose at first yellow green, then deep green 2.5 to 4 $\mu$  in diameter, becoming rough. Habitat: The air of north Italy. This species was recognized as near *P. rugulosum* Thom., differing only in minor details.

406. *P. rugulosum* var. *atricolum* (Bainier?) Thom., n. var.

Colonies upon Czapek's solution agar small, restrictedly growing, about 1 cm. in diameter in ten to twelve days, gray-green, to gray with outer area white or nearly so and a submerged band perhaps 1 mm. wide; reverse uncolored in Czapek plates, becoming orange upon corn meal; conidiophores 100 to 200 $\mu$  long; penicillus variously monoverticillate, biverticillate and symmetrical or with metulae and sterigmata in the same verticil; sterigmata 8 to 9 $\mu$  in length, lanceolate; conidia about 3.5 by 2 $\mu$ , rough.

Culture no. 4640.439 as received in the Bainier collection was labeled *P. atricolum* and is made the type. The relations among the many variant strains assigned by us to *P. rugulosum* have not been worked out fully. Strains with white margin and colorless reverse as in Bainier's culture are fairly common. Most of them show tardily a development of the orange colors in reverse and the yellow aerial hyphae of *P. rugulosum*. This varietal name may be justified by these differences.

407. *P. chrysitis* Biourge. Monogr. La Cellule 33: fasc. 1, p. 252; Col. Pl. XI, Cart. 410; Pl. XIX, fig. 112. 1923.

Colonies in wort gelatine, restricted in growth, undulate-plicate; upon Czapek's solution agar partly funiculose forming a close textured felt of fine hyphae presenting a surface almost velvety in appearance, with marginal area showing abundant hyphae richly covered with yellow granules and many mycelial hyphae filled with orange coloring matter;

conidial areas deep dark green to olive green to purple brown in age; reverse orange, with margin golden yellow to orange; agar colored a lighter shade; conidiophores  $100\mu$ , sometimes up to  $200\mu$  long by  $2.8$  to  $3\mu$ ; penicillus about  $20\mu$  long, with walls smooth, figured as a symmetrically biverticillate mass; metulae  $8$  to  $10$  by  $2$  to  $3\mu$ , commonly in fours; sterigmata  $9$  to  $12$  by  $2$  to  $3\mu$ , in threes, fours, or fives; conidia rough, ovate,  $3.5$  by  $2.8\mu$ , with connective persistent.

Biourge's type no. 410 (our no. 4733.32) was received in September, 1927, and is closely related to *P. rugulosum* but shows a slightly lighter orange color in reverse than Biourge's strain of *P. rugulosum* no. 4733.113. The description furnishes little justification for separating it unless the aggregate of strains we have included in *P. rugulosum* are to receive names enough to cover all the quantitative variations in color production encountered.

408. *P. crateriforme*, Gilman and Abbott. Iowa State College Jour. Sci. 1: no. 3, p. 293, fig. 28. 1927.

Emended description: Colonies on Czapek's solution agar, velvety or with little aerial mycelium, restricted in growth, with conidial area deep dark green to almost black in age, uneven in surface, from dense irregular masses of conidia becoming  $300$  to  $400\mu$  deep; in other cultures the rough or irregular areas develop as ascending ropes almost coremia, linking the species with the coremiforme series; inner marginal band varying from citrine to yellowish green then an outer uneven or crenulate white margin  $1$  to  $2$  mm. wide; overgrowth of yellowish hyphae occasional; mycelium consisting of close-woven delicate hyphae penetrating rather deeply into the agar, tearing easily; reverse at first colorless or with the dark green of the conidial area showing through, later partly or in zones orange red (Corinthian red of Ridgway) especially near the margin; odor faint or indefinite; drops not seen; conidiophores mostly very short, up to  $100$  or  $150\mu$  by  $3$  to  $3.5\mu$ ; penicillus a close-packed verticil of metulae with close-packed sterigmata forming a v-shaped mass supporting chains of conidia parallel or slightly diverging at first but forming dense masses as seen in mature cultures; metulae up to  $12$  by  $3$  to  $3.5\mu$ ; sterigmata  $8$  to  $10$  by  $2\mu$  acuminate; conidia elliptical to fusiform at first, later subglobose up to  $3.5$  to  $4$  by  $3$  to  $3.5\mu$ , or  $3.5$  to  $4\mu$  with dark outer wall showing more or less regularity.

Emendations and expansion of diagnosis made from Gilman and Abbott's type culture our no. 4894.13. Also no. 5027 B from R. E. Wean, Lafayette, Ind.

*P. purpurogenum* series. Reverse predominantly in bright red colors; includes no. 409 to 413.

409. *P. rubrum* O. Stoll. Beitr. z. morph. u. biol. Char. Penicillium Wurzburg 1904, p. 35, Taf. I, fig. 7, Taf. III, fig. 3, Taf. IV, fig. 4. See also Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, p. 39, fig. 7, 1910.

Colonies upon Czapek's solution agar, slowly growing, small, restricted, not widely spreading, green modified by various mixtures of hyphae studded with yellow to reddish granules; close woven mycelium with surface velvety in appearance and reverse red; substratum also colored in age; conidiophores commonly 15 to 30 by 3 to 3.5 $\mu$ , arising directly from substratum or from creeping, looping, felted hyphae. Metulae in a close verticil, with sterigmata 10 to 13 by 2 to 3, and chains of conidia 3.4 by 2 to subglobose up to 3.3 in a dense column; conidia elliptical 3.4 by 2 $\mu$  to subglobose 3.3 to 2.5 $\mu$ , yellow to dark green.

Cultures: By Stoll, by Thom but not maintained in collection.

Biourge's (Monogr., pp. 172-174) culture labeled *P. rubrum* (our no. 4733.111) is an entirely different organism and closely related to his own *P. rubens* with which we have placed our notes upon it.

410. *P. sanguineum* Sopp. Monogr., pp. 175-176; Taf. XIX, fig. 138; Taf. XXIII, fig. 24. 1912.

Colonies on meat-peptone-sugar-gelatine, with restricted central green conidial area, then a broad, deep zone of mycelium, at first (see Sopp's figure) yellow then red, and the substratum quickly colored bright blood red; reverse red from the first; hyphae moderately coarse; gelatine only slightly liquefied; odor aromatic and alcoholic; conidiophores and penicillus typical of the verticillate series; conidia 3.5 by 2.5 $\mu$ ; perithecia not found.

Species found upon earth and leather. Cultures grew best at 20° with minimum at 3° and maximum at 35°C., and moderately well upon the other media tested.

Conidia remained viable more than three years.

Sopp's description and figure show that this was some member of biverticillate group; no one can be absolutely certain of the strain but in our own soil cultures we have many times obtained a form carried as nos. 4917.7 and 4927 in our collection which complies nearly enough with these characters to propose the use of the name: Colonies upon Czapek's solution agar showing well differentiated conidial zones often

with sterile separating zones, with conidial masses a shade of dark yellow green near Andover green (Ridgway XLVII.); zonation often obliterated in age and sometimes wanting at any stage; overgrowth of orange hyphae more or less evident in many cultures in age; reverse a shade of orange red, near Corinthian red (Ridgway XXVII.) without showing yellow shades; conidiophores 50 to 100 $\mu$  by 4 $\mu$ ; with walls smooth, penicillus biverticillate, sometimes with the third verticil more or less unsymmetrical, with long chains of conidia parallel, or the chains from the single verticils of sterigmata more or less adherent into columns and the columns forming more or less continuous masses up to 400 $\mu$  thick which break off in crusts when the container is tapped or struck; metulae up to 10 to 12 by 2 to 3 $\mu$ , in compact verticils; sterigmata acuminate up to 10 to 12 by 2 to 2.5 $\mu$ , in compact verticils; conidia elliptical to subglobose, 3 by 2.5, to 3.5 by 3 $\mu$  or 3 to 3.5 rarely 4 $\mu$  when subglobose, green, becoming dark green in mass.

Strains of this type are exceedingly common in cultures from soil at Washington, D. C., by members of the laboratory of Soil Microbiology.

411. *P. variabile* Sopp. Monogr., pp. 169–171; Taf. XVIII, fig. 124; Taf. XXIII, fig. 27. 1912.

Colonies on meat-peptone-sugar-gelatine showing a wide range of color in red and green shades, and in reverse from light to very dark red almost black; mycelium orange to rose or carmine red, on some substrata in shades of green and yellow green; substratum from pink red to deep blood red; hyphae comparatively fine or delicate; odor suggestive of the bark of Aspen (*Populus tremuloides*?); conidiophores rather coarse, stiff, septate, brownish, thick walled, swollen somewhat at apex and producing a verticil of 2–6 metulae often with one or more superposed verticils produced by prolongation of the main stalk; sterigmata narrowly cylindrical ("needle-like"), 8 to 20 in the verticil; conidia elliptical, fusiform 3 by 4 $\mu$ , with a tendency to become adherent in a ball, like *Gliocladium*, figured as chains more or less adherent into columns; perithecia not found.

Species found on rotting paper in earth. Cultures grew best at 20°, with a minimum at 5° and maximum of 35°C. Colonies grew well on Sopp's gelatine and agar, wort, potato and rice, less well on milk, urine and broth.

Conidia remained viable more than three years.

A culture, no. 5017.80a, coming as a contaminant in and parasitization of a culture of *A. tamarii* from the Federated Malay states, showed a

varying mixture of yellow green and ultimately red colors which might account for Sopp's name *P. variabile*. Its conidia were smooth or nearly so—otherwise the structures were those of *P. rugulosum*.

412. *P. purpurogenum* Stoll. Beitr. a. Morph. u. biol. Char. Penicill. Wurzburg (1904), p. 32, t. I, fig. 6, t. III, fig. 2. See also Thom, U. S. Dept. of Agr., Bur. Anim. Ind. Bul. 118, p. 36, fig. 5. 1910 and Mycolgia 7 (1915) No. 3, p. 134 to 142.

Colonies in conidial areas green with more or less tardily developing hyphae studded with yellow granules, as interlacing fibers and tufted over growths; reverse through yellow to orange tints to red; conidiophores 100 to 300 by 3 to 3.5 $\mu$  arising directly from the substratum or from a floccose web of superficial hyphae; penicillus symmetrically biverticillate; sterigmata 10 to 12 by 2.5; conidia 3.4 to 3.8 by 2 to 2.5, with walls often very delicately granular, in chains more or less diverging to tangled in mass. Culture: by Stoll (origin of culture attributed to Fleroff), by Thom, No. 17 from Stoll's culture through Westerdijk, and many contributed and independently isolated cultures closely related.

There appear to be many strains with the general morphology and reactions of *P. purpurogenum*. These differ somewhat in colony characters, depth of mass, abundance of conidia, rate of color development and especially in rate of change from yellow to red. Separation into species or even varieties within this series is more or less precarious. We have, therefore, presented *P. purpurogenum* as a convenient aggregate leaving close description of its components for future researches.

- 412a. *P. purpurogenum* Fleroff-Stoll. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 235-237; Col. Pl. VII, Cart. 54B; Pl. XI, fig. 66. 1923.

*Synonym.* *P. sanguineum* Sopp fide Biourge.

Colonies on wort gelatine at first olive green with margin for a long time yellow, in age brown; coremia none; reverse purple red with a yellow margin, in age cinnabar or deep blood red; agar red; odor none; conidiophores 2 to 3.5 in diameter, all walls smooth; penicillus figured as about 25 $\mu$  in length when a simple verticil of metulae, 45 to 60 $\mu$  in length when a secondary verticil is developed from a lower node; metulae about 12 by 2.5 $\mu$  commonly in fours; sterigmata 10 to 17 by 2.5 to 3.5 $\mu$  in groups of 2 to 4; conidia elliptical 3.4 to 4.4 by 2 to 3 $\mu$ .

Biourge's no. 54B (our no. 4733.100 not Thom no. 17) when grown

in Czapek's solution agar, produced colonies with margin submerged, then a velvety zone about  $200\mu$  deep, then with central area floccose, conidial areas dull green; reverse purple reddish, not abundantly colored; measurements approximately as in Biourge.

Biourge's discussion is cited in full as illustrating the variations encountered among these species and his form of description applied to material differing little from ours.

3. *P. purpurogenum* var. *rubri-sclerotium* Thom, Mycologia 7: 141-142, fig. 1. 1915.

Differs from typical description of the species in the production of dark-red sclerotia on the surface of the substratum and in the well marked zone of yellow at the margin of the growing colony. It appears to be a soil fungus with well marked characters.

Type no. 2670 of Thom's collection, isolated from corn grown in Virginia in 1912. Several other strains of this variety have been studied. No. 4756 was isolated as the cause of red spots in moistened print paper in the United States Bureau of Engraving and Printing where it had rendered large masses of paper worthless for printing purposes; no. 4016 was found in the laboratory upon jelly in 1914; no. 2647 was contributed by Miss Dale from English soil and is cited in her paper as *P. rugulosum* Thom.

This culture in laboratory transfers over a period of more than fifteen years has lost to a considerable extent the details of colony structure found when it was freshly isolated. Sclerotium production has also been suppressed or lost. Similarly no. 4756 has lost the sclerotium producing power characteristic of the original isolation. No. 2670 was selected by Dr. May at the gluconic acid producing species of the May, Herrick and coworkers series of papers. (See Gluconic Acid Chapter VIII.)

*P. luteum* series. Floccose colonies in yellow and green with reverse predominantly in yellows or tardily red; includes nos. 415-418.

In the sense already described cultures are constantly encountered in which the conidial forms vary only in minor detail from a general or type description. It is doubtful if description of species among them with our present knowledge has any value but certain species already described are included here.

Nonascosporic strains of the *P. luteum* series have been collected from many sources. Biourge's no. 368 (our 4733.85) as described appears to have been derived from our no. 11 which is still regularly

ascosporic. As received from Biourge, this is a floccose colony, white, with slow development of areas or zones gray from ripening conidia. In reverse the cultures become a yellowish orange shade near pinkish cinnamon (Ridgway XXIX.15). It would appear probable from Derx's work that in handling this culture one of the haplonts necessary for ascospore production has been eliminated by successive selective transfers. In our own exchanges a strain identical to no. 4733.85 has been received from C. M. Tompkins (no. 5042.87) at Logan, Utah, as obtained from decaying sugar beets. In handling many thousands of petri dish cultures from soil, food, and miscellaneous substrata, many strains of this group have been seen varying from forms which produce abundant green conidia covering the whole mycelium with only traces of yellow sterile hyphae, to colonies predominantly consisting of sterile yellow hyphae with scattered green areas and finally forms in which most of the usual characters of the group are suppressed leaving only few and scattered biverticillate penicilli to trace the relationship.

Systematic study of the presence of so-called plus and minus strains in the sense of Blakeslee, of the combinations necessary to produce perithecia, and full description of the ascospores obtained, may be expected eventually to clear up the uncertainties in this group. Schwartz and Derx have made some progress. Further reports may be expected.

415. *P. aureolimbium* Zaleski. In Bul. Acad. Polonaise, Sci: Math. et Nat. Ser. B, 1927, pp. 481, 482; Taf. 53; Zaleski no. 563.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 26 to 30 mm. in diameter in twelve days, liquefying the gelatine quickly and completely, with central area characterized by cerebriform wrinkles 1 to 2 mm. high and 1 to 2 mm. wide, which become radiate in the outer areas; marginal area velvety, zonate with the outer zone or "fimbria" 1 mm. in width; in color conidial areas at first blue green 378B, to 396, 372, 366, becoming later green 346, 347, 322, 318; reverse in orange yellow shades such as 171, 166, 151, 126; odor rather foetid, strong, similar to rotting potatoes; conidiophores varying 100, commonly 150 to 300, less commonly 500, by 2.5 to  $3\mu$ , straight or flexuous, simple or rarely branched, with apices usually inflated, with all walls smooth; penicilli about 18 to  $25\mu$  in length; metulae about 12 to 15 or even 18 by 2.5 to  $3\mu$ , in groups of 3 to 6, with apices variously enlarged or inflated, equal or unequal in length and symmetrically or asymmetrically arranged in the verticil; sterigmata about 8 to 9 by 2.2 to  $2.5\mu$ , in verticils of 6 to 10 with short tubes; conidia 2 to 2.5 or even

2.8 $\mu$ , smooth, more or less globose, showing distinct connectives in the chains.

Habitat: Species isolated from earth under pines in the hills "Poroniec" in the Tatry mountains in Poland.

Zaleski doubtfully identified this species with *P. aurifluum*. Biourge decided the species to be new but related to *P. aurifluum* from which it is distinguished by the marginal zone of submerged orange mycelium seen in reverse and giving the name *aureo limbum*. Zaleski classes it in "Biverticillium Dierckx Subsec. 4. Cerebriformiter-radiate-undulata."

Culture no. 5010.3 received from Baarn in July, 1928, does not give colonies complying with Zaleski's description. Probably his green form

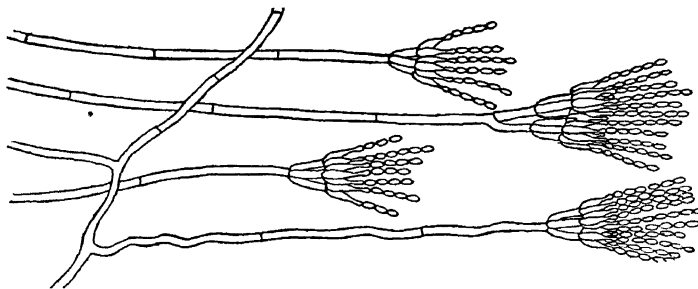


FIG. 82. *P. citricolum* Bainier and Sartory (part of Bainier and Sartory's plate): Detail of penicillus, conidiophore and its origin.

has been entirely replaced with a floccose white species. From the description alone we are compelled to believe he had one of this series in culture.

Zaleski was clearly unfamiliar with the large number of yellow green species of this group.

416. *P. citricolum* Bainier and Sartory. Bul. Soc. Mycol. France 28: 276-279, Pl. XIII, fig. 1, 2. 1912. (See fig. 82.)

Colonies on licorice sticks with conidial areas yellowish green (C.d.C. 313, then 263-267-257), on some media blue green; reverse and medium citrine yellow; conidiophores 2 $\mu$  in diameter, figured as septate, more or less sinuous, rather long and arising from creeping or ascending hyphae; penicillus consisting of a verticil of 4 to 6 closely crowded metulae about 8 $\mu$  long each bearing 3 to 6 sterigmata; conidia oval 2 $\mu$  in diameter (apparently in short axis—C. T.).



Species found upon an orange peel. Cultures grew best at 26° to 28°C. They liquefied gelatine producing yellow pigment.

The description given places this form in the *P. luteum* section of the biverticillate group but lacks the necessary details to insure any closer identification.

As a possible identification we may cite no. 4917.6 with abundant aerial mycelium yellow (fringed with yellow granules) as a background for deep green conidial masses borne upon branches or conidiophores of varying length mostly arising from aerial hyphae.

417. *P. luteum* (?) after Sopp. Monogr., pp. 173-175; Taf. XXIII, fig. 23. 1912.

Colonies upon meat-peptone-sugar-gelatine with conidial areas restricted but dark gray green or dark olive green upon a pale yellow mycelium, growing as a heavy colony along the line of inoculation but not spreading widely over the substratum; reverse reddish or pale flesh color, becoming black upon some media; odor strong alcoholic and aromatic; conidiophores short, uniformly producing a biverticillate penicillus, with metulae comparatively long, numerous, cylindrical; sterigmata rather long and flask-shaped; conidia partly elliptical, partly globose 2 to 3 $\mu$  in long axis; perithecia not found.

Species found upon earth, leather, and cotton. Cultures grew well at 20°, with a minimum of 5° and a maximum of 33°C., with fairly good growth upon the common media tested.

Conidia remained viable more than three years.

There is very little difference between this and *P. sulfureum* of Sopp hence it is scarcely identifiable.

418. *P. sulfureum* Sopp. Monogr., pp. 172-173, Taf. XVIII (corrected [C. T.] XVII), fig. 120; Taf. XXIII, fig. 22. 1912.

Colonies upon meat-peptone-sugar-gelatine with sulphur yellow to reddish yellow mycelium and deep green or olive green conidial areas, with a tendency toward coremium formation and to produce heavy masses of conidia which break off readily; mycelium floccose, spreading slowly over the substratum, with a fibrous appearance ("filzig"); reverse from sulphur yellow to red; substratum mostly colored reddish to blood red; conidiophores comparatively stout, septate, rarely branched, uniformly biverticillate, forming dense conidial masses, breaking away easily; metulae figured as fairly slender, uniform and close packed in the verticil; sterigmata shown as long narrow and almost

parallel in the verticil; conidia small, elliptical  $2.5$  to  $3\mu$  in long axis; perithecia not found.

Species found upon a rotten apple where heavy coremia were produced; cultures grew best at  $20^{\circ}$  with a minimum of  $3^{\circ}$  and a maximum of  $33^{\circ}\text{C.}$ , and grew well upon the common media tested—wort, rice, milk, potato broth, gelatine and agar. Conidia remained viable more than 3 years.

Although no one has surely found this species we have had several of the *P. luteum* series which refused to produce ascospores, showed large areas of sulphur yellow mycelium and occasionally structures suggesting coremia.

*Section 5. Biverticillata-miscellanea.* Species included are not obviously closely related to the usual biverticillate species, but agree with them in the general morphology of the conidiophore.

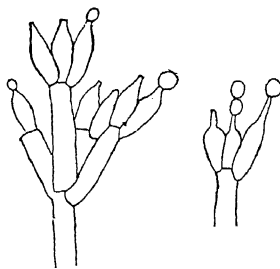


FIG. 83. *P. Braziliensiense* Thom: Detail of penicillus

Species such as *P. citrinum* Thom, *P. corylophilum*, *P. steckii* which we have placed with the velutinous divaricate series might almost equally well be placed here.

*Conidial areas colorless to cream showing no shade of green.*

420. *P. Braziliense* Thom, n. sp. Type no. 4707.759 I from Dr. Da-Fonseca at Rio de Janeiro, Brazil. See fig. 83.

Colonies white or very faintly tinged, velvety, spreading, broadly, not over  $200\mu$  deep, slowly and incompletely zonate in age; reverse (four weeks) yellow to olive buff or a dirty yellow orange mixture; drops not seen; odor none; conidiophores  $100$  to  $200\mu$  long ascending rather than erect, by  $3\mu$  to  $4\mu$ , with walls pitted or rough; penicilli variously branched only partly biverticillate; metulae  $16$  to  $20\mu$  en-

larged at apex unequal in the verticil; chains tangled—breaking up in mounts; sterigmata 13 to 16 $\mu$ ; conidia about 3 $\mu$ —very pale with more or less internally granular appearance to thin walls.

This culture as received from Dr. DaFonseca at Rio de Janeiro has been seen but once. It is given because white forms with partly biverticillate penicilli do occur and until further studied would fall at this place in the scheme of classification.

*Conidial areas showing some shade of green.*

421. *P. namylowski* Zaleski. In Bul. Acad. Polonaise Sci: Math. et Nat. Sér. B, 1927, pp. 479, 480; Taf. 52; Zaleski no. 1430.

Colonies in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing, becoming 24 to 28 mm. in diameter in twelve days, liquefying the gelatine only slightly, with surface uneven, velvety, rarely indistinctly zonate near the margin; central area showing deep cerebriform wrinkles which become radiate outwardly; margin 1 to 1.5 mm. wide during the growing period; in color conidial areas green 346, 347 becoming dark orange brown shades such as 148 in age; reverse, yellow green, to yellow shades such as 0296, 277, 171, odor strong, offensive; conidiophores 70 to 150 or even 300 by 3.5 to 4 $\mu$ , with apices enlarged usually to 5 to 6 $\mu$ , straight or flexuous, simple or rarely branched, with walls smooth; penicilli 20 to 30 $\mu$  when simple, 40 to 50 $\mu$  when a branch is present; branches rare, when present 20 to 25 by 3 to 4 $\mu$ , with apex enlarged to 6 $\mu$ ; metulae about 14 to 20 by 3 to 4 $\mu$  with apices vesicle-like, equal or unequal in length, in symmetrical or asymmetrical verticils of 3 to 6; sterigmata about 9.5 to 11 by 3 to 3.5 $\mu$ , commonly 6 to 10 (or even 15) in the verticil, with short heavy tubes, crowded in the verticils and straight or incurved conidia 2.5 to 4 $\mu$  by 2.5 to 3.0 (even to 3.5 $\mu$ ), smooth, long, ovate to subglobose, showing distinct connectives in the chains.

Habitat: Species isolated from earth under pines in square "652" of the forest "Puszcza Bialowieska" in Poland.

Zaleski notes that Biourge did not know of a *Biverticillium* with foetid odor, but was not able to eliminate it from the group. Zaleski himself put it among his "*Biverticillium* Dierckx Subsec. 4. Cerebriformiter-radiate-undulata." Our cultural notes follow: Type strain growing equally well at 30 and 20 C. Colonies upon Czapek's solution agar growing as a very thin colorless submerged mycelium with few pin head clusters of penicilli thinly scattered; single penicilli visible only

under a lens; upon wort or dextrose agar wrinkled closewoven velvety olive in color; reverse uncolored; drops, none; odor, strong, penetrating; conidiophores from very short to  $200\mu$  long by  $4\mu$ , with walls pitted, granular or rough with walls showing rough as seen by viewing the whole colony under low magnification; penicilli with metulae borne symmetrically or more or less unsymmetrically 10 to  $15\mu$  long, with conidial chains parallel, adherent in groups or twisted forming one or more columnar masses up to 300 to  $400\mu$  long; sterigmata 10 to 12 by 2 to  $2.5\mu$ ; conidia cut off first as a chain of cylindrical segments 2 to 3 by  $1.5$  to  $2\mu$  then slowly enlarging and rounding up toward 4 by  $3\mu$ .

Culture no. 5010.16 received from Baarn in July, 1928, appears to be type. Several strains identical or nearly related to this species were isolated in this laboratory during the same month from soil upon the Arlington Farm, United States Department of Agriculture, and had been fully described as a species at the time Zaleski's culture was first studied by us.

422. *P. tardum* Thom. (Fig. 84).

*P. elongatum* Bainier, Bul. Soc. Mycol. France 23: 17-18; Pl. V, fig. 1-7. 1907.

Not *P. elongatum* Dierckx, 1901, q.v.

Colonies upon licorice sticks, at first almost white, then pale blue, and slowly changing to gray green; conidiophores long, slender and figured as bearing terminal penicilli on the main axis and upon various long diverging branches irregularly disposed along the ascending or creeping fertile hypha; penicillus figured as symmetrically biverticillate, with 3 to 8 metulae each bearing 3 to 5 sterigmata about as long as the metulae; with metulae and sterigmata forming a graceful closely packed mass; conidia oval about  $2.8$  by  $1.4$  to  $1.6\mu$ , swell, become globose only before germinating and emit one or two delicate tubes.

Bainier reported his species from twigs of dead wood. Our culture no. 4640.444 from the Bainier collection appears to be correctly named, while no. 4640.425, and no. 4640.420 from the same collection were also this species; no. 4177 from Hesler, no. 4658.7.1, and no. 75.2 from Putterill in South Africa appear to be identical with them. From study of these forms, we have emended Bainier's description as follows: Colonies on Czapek's solution agar, slow and restricted in growth, green, with a submerged zone 3 to 4 mm. broad; reverse and agar not colored; odor none; vegetative hyphae slender; conidiophores slender, 300 to  $400\mu$  by 2 to  $2.5\mu$ , bearing a symmetrical verticil of metulae and sterigmata

characteristic of the group; metulae unequal in the verticil but about 8 to 10 by  $2\mu$ ; sterigmata up to 8 by  $2\mu$ ; conidia elliptical mostly up to  $2.5$  to  $3\mu$  in long axis, less often subglobose, in slightly diverging or fairly closely arranged but not adherent chains, forming masses up to  $160\mu$  long and broadening to  $100\mu$  at the apex.

Biourge places the species in his *Biverticillium* (Monogr. p. 254). It does not produce the red and yellow colors in the substratum so characteristic of the principal sections of the group. Since Dierckx published the name for a different species in 1901, we have given the name *P. tardum* because of its slow development in our standard substratum—Czapek's solution agar. No. 4920.8 from soil in the Arlington farm near Washington reproduces the morphology of *P. tardum* but grows more freely than Bainier's culture as received from France. No. 5034.45 from Birkinshaw appears to be this species.

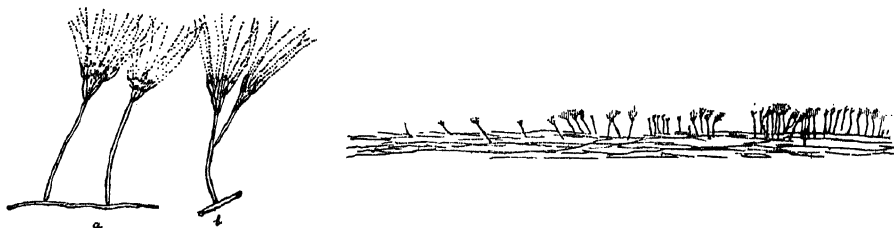


FIG. 84. *P. tardum* Thom: a, b, habit sketches; c, diagrammatic radial section of developing colony upon Czapek's solution agar.

423: *Paecilomyces erectus* Demelius. Verhandl. Zool.-Bot. Gesellsch. Wien. 72: 78-79. fig. 7, 8, (1922) 1923.

Not *Paecilomyces* Bainier.

Colonies very small, round or irregular, 0.5 to 3 mm. in diameter, gray green (caesius) (C.d.C. 346) velvety; vegetative hyphae 3 to  $4.8\mu$  in diameter; conidiophores 120 to  $240\mu$  by  $2.4$  to  $3.6\mu$ , erect, septate, hyaline simply branched or frequently forked twice or three times, each tip bearing a verticil of 2 to 5 sterigmata; sterigmata fusoid 17 to 24 by  $2.2$  to  $3\mu$ , in verticils of 2 to 5, figured (fig. 7) with long beak; conidia single or in short chains ellipsoid  $3.6$  to  $5$  by  $1.8$  to  $2.4\mu$ , rarely  $7.2$  by  $3.6\mu$ .

Species was reported as found on a potato from Moravia at Vindobon, 1915, and as differing from other species of the genus in its erect, richly branched conidiophore.

Bainier's *Paecilomyces* is based upon an organism which is never

green and differs markedly in habit from this form which must be placed tentatively in the miscellaneous section of doubtful members of Biverticillium. No change of name is proposed. When some one completes the study of this form on standard media, a more accurate identification may be anticipated.

If the colors of the conidial areas contain no real green element, the describer may have been justified in placing her species in *Paecilomyces* but it differs so greatly aside from color as to raise a preponderating doubt.

*P. piscarium* Westling. Arkiv för Botanik 11: 54, 86-88, fig. 13 and 55. 1911. (Compare fig. 85.)

Colonies in prune gelatine floccose, with conidial areas bluish or pale blue green (C.d.C. 422, 387, 383, 384, 393) forming the center with mar-

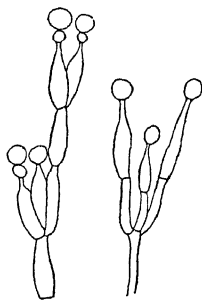


FIG. 85. *P. piscarium* Westling: Anomalous forms of penicillus characterize the species.

gin broad, somewhat woolly white, the whole colony becoming dark gray in age; reverse uncolored or pale yellowish; gelatine liquefied; odor none; abundant drops of colorless fluid observed upon the growing colony; conidiophores up to 1 mm. by  $3.2$  to  $5\mu$ , with walls smooth, arising from creeping or submerged hyphae, or much shorter when arising from aerial hyphae, penicillus figured as biverticillate and observed in Westling's culture as irregularly biverticillate with many reduced or partial verticils; metulae  $10.5$  to  $14$  by  $3$  to  $4.8\mu$ ; sterigmata  $8$  to  $9$  by  $2.6$  to  $3\mu$ ; conidia smooth, elliptical to ovate,  $3.4$  to  $4.2$  even to  $4.6\mu$  by  $2.8$  to  $3.4\mu$ .

Species found in emulsion of cod liver oil, with "pyrophosphas ferrico-ammonicus," the salt being apparently essential for development of the

mold. Calcium-oxalate crystals, sphaerocrystals, needles and rhombic forms were observed among the hyphae

Cultures grew well at 30° to 31°C., grew poorly upon 10 per cent tannin solution, changed litmus gelatine to red and grew well upon malt extract gelatine, potato, bread, and Maranta-starch. Upon milk the mycelium produced became yellow but few conidia were seen. No growth appeared upon citric acid solution.

Westling's culture (no. 2549) has been kept for many years and shows a member of the section of the biverticillate series in which yellow and red color production has been suppressed or greatly reduced to appear only occasionally and slightly.

Biourge (Monogr. La Cellule 33: fasc. 1, pp. 190-191; Col. Pl. XI, Cart. 376; Pl. XVIII, fig. 107, 1923) discusses as *P. piscarium* his no. 376 (our no. 4733.97), which is apparently the one (no. 2549) sent to him by Thom. It is assigned by Biourge to the group with *P. chrysogenum* arbitrarily while he appears to believe it belongs with *Spicaria*. Culture no. 4733.97 grown on Czapek's solution agaf reproduces Biourge's figure fairly well. He shows the penicillus as consisting of a verticil of branches or metulae very irregular in length producing either verticils of few sterigmata or growing out into hyphae but fails to show the typically long lanceolate sterigmata. The conidia are mostly 3 to 3.5 by 2.5 to 3, less commonly subglobose 3 $\mu$ , and delicately roughened.

425. *P. Miczynskii* Zaleski. In Bul. Acad. Polonaise Sci.: Math. et. Nat. Ser. B, 1927, pp. 482, 483, 484; Taf. 46, 53; Zaleski no. 574c.

Conidia in neutral Raulin with 10 per cent gelatine in petri dishes, slowly growing becoming 32 to 35 mm. in diameter in twelve days, liquefying the gelatine quickly and completely; surface velvety-sub-lanose, azonate, with marginal area plane or slightly elevated, the center cerebriform wrinkled and depressed, passing outward as radiate wrinkles; surface growth consisting of a dense lanose mass of white mycelium tardily showing a faint bluish green and later green in the central area, tending again to be overgrown with white mycelium, only locally showing gray green conidial areas; reverse and liquefied gelatine in yellow shades such as 221, 191, 178A, 216, 211; odor weak or none; conidio-phores 300, 400 to 800 or even 1000 $\mu$  by 2.5 to 3.5 $\mu$  commonly enlarged at the apices, simple or rarely branched, straight or flexuous, all walls smooth; branches rarely encountered, usually unequal, distant, 15, 20 to 30 or even 36 $\mu$  by 2.5 to 3 $\mu$ , sometimes 2 or 3 at the node; penicilli

18, 22 to 28 or even  $35\mu$  long; metulae about 12 to 18 or even  $24\mu$  by  $2.5$  to  $3.5\mu$ , commonly enlarged upward, usually 4 to 8 in the verticil in which the units are unequal in length and symmetrically or rarely unsymmetrically arranged; sterigmata about  $8.5$  to  $9.5$  by  $2.0$  to  $2.5\mu$ , commonly 6 to 10 in the verticil, with short tubes; conidia 2 to  $2.5$  or even  $3\mu$ , smooth, subglobose or occasionally globose.

Habitat: Species isolated from earth under conifers in "Poroniec" in the Tatry Mountains in Poland.

Zaleski classes it in "Biverticillium Dierckx, Subsec. 4. Cerebriformiter-radiate-undulata." Our cultural notes follow: Type strain growing better at  $20^\circ$  than at  $30^\circ\text{C}$ . or higher. Colonies upon Czapek's solution agar at  $20^\circ\text{C}$ ., about 35 mm. in diameter in fourteen days, cottony floccose, deeply wrinkled, white then slowly gray (very slightly greenish), with white deeply floccose marginal area and fimbriate submerged mycelium at the very edge; mycelial hyphae very delicate; reverse uncolored, yellow to greenish yellow (Ridgway XXXI) chrysolite green to seafoam green; conidiophores unequal in length, very slender, about  $2\mu$  in diameter; penicilli variously monoverticillate, biverticillate, or a main axis and one branch with biverticillate penicilli at different levels; sterigmata 10 to  $12\mu$  long; conidia about  $2.5$  to 3 by  $2\mu$ .

Culture no. 5010.15 received from Baarn in July, 1928, appears to satisfy Zaleski's description of his type; it suggests Sopp's *P. canescens* in several characters but has been tentatively kept in the biverticillate group on account of the morphology of the penicillus.

426. *P. Rolfsii* Thom, n. sp. Type and description no. "32" in U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118.; p. 80, 81, fig. 36, 1910. (Fig. 86.)

Colonies upon milk-sugar-gelatin and potato or bean agar gray-green; floccose, but with aerial part mostly long conidiophores and few vegetative hyphae, slightly yellowish to pronounced salmon color below; broadly spreading; developing elliptical to globose sclerotia 150 to  $200\mu$  in diameter at the surface of the substratum in 2 to 3 weeks, conidiophores 200 to  $500\mu$  by 3 to  $4\mu$ ; penicilli verticils of 3 to 5 branches 10 to  $17\mu$  by 2 to  $3\mu$  rarely secondary verticils each bearing a dense verticil of sterigmata, 8 to  $10\mu$  by  $2\mu$  producing long, parallel, or slightly divergent chains of conidia; conidia elliptical or fusiform,  $3.5$  to  $4\mu$  by 2 to  $3\mu$ , green, granular within, smooth, swelling in germination to  $6\mu$  and producing from one to several germ tubes. Colonies slowly liquefy milk-sugar-gelatin and produce purple or neutral colors in litmus media.



Sent by Prof. P. H. Rolfs from Miami, Fla., upon portion of pineapple, March, 1905.

Color gray-green, with scattered white to pink sclerotia; reverse, sulphur-yellowish to pronounced salmon; color in media reddish or yellowish in special cases, others none. Odor, none.

Fifteen per cent gelatin in water, typical; liquefaction, none in fifteen days, later very slow liquefaction; litmus reaction neutral, leaves both acid and alkaline media purple-blue. Potato agar and bean agar, typical, slightly thinner than gelatin cultures, gray-green without sugar, clear green with cane sugar. Potato plugs, typical, transpires yellow drops which become very dark yellow (balsam). Raulin's fluid, good colonies becoming rosy below. Cohn's solution, small colonies, fluid slightly yellow.

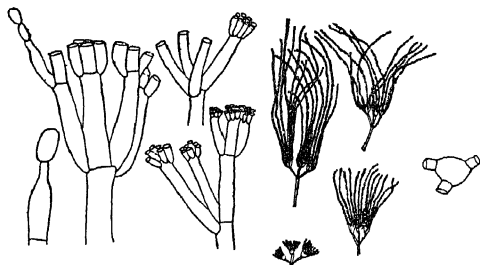


FIG. 86. *P. Rolfsii* Thom from 1910 paper: Anomalous biverticillate penicilli but with relationship to this group predominating.

Synthetic fluids (Dox's), carbon supplied as: Cane sugar, grew in solutions up to 30 per cent with acid reaction. Lactose 3 per cent, very slow growth of small characteristic colonies. Lactic acid 0.9 per cent, good growth, light green. Levulose 3 per cent, very slow-growing but heavy colonies. Galactose 3 $\frac{1}{2}$  per cent, typical. Glycerin, very small colonies. Butterfat, typical colonies.

Milk, curdling (0.25 per cent calcium chlorid added) very slow; digestion, very slow; color in milk, none.

At 37°C., grew more rapidly than check at 20°C.

427. *P. lagerheimi* Westling. Arkiv för Botanik 11, pp. 55, 110-112, fig. 25, 66a and b. 1911.

Colonies in prune gelatine, floccose, green (prasinus C.d.C. 346 then 347, 342) then gray green (322, 348) and finally brownish in old cultures:

with broad white margin; reverse uncolored or brownish; gelatine liquefied after a month; odor weakly aromatic; transpired crops clear, small; conidiophores arising from aerial hyphae, smooth, from 200 up to 800 $\mu$  by 3 to 4.6 $\mu$ ; penicillus 60 to 150 $\mu$  long, figured as either monoverticillate or biverticillate but described as if comparable to other species with more complex branching of the conidial apparatus; metulae 10 to 18 by 3 to 4.5 $\mu$ ; sterigmata 7.5 to 10 by 1.8 to 2.6 $\mu$ ; conidia elliptical or oblong, smooth 2.7 to 3.8 even to 4 or 4.8 $\mu$  by 2 to 2.8 $\mu$  or even 3.8 $\mu$ .

Species found on fruits of *Ribes grossularia* L. It produced a poor colony at 30° to 31°C., but grew well upon the media regularly used by Westling, malt-extract-gelatine, potato, bread, plum agar, maranta starch and milk. The type culture (no. 2544) was received from Westling but has long been lost from our collection.

427a. *P. lagerheimii* Westling. In Biourge, Monogr. La Cellule 33: fasc. 1, p. 198; Col. Pl. III, Cart. 380; Pl. V, fig. 30. 1923.

Colonies in wort gelatine, deeply lanose, yellowish white, coremia none; reverse orange yellow, odor indefinite; conidiophores 1.2 to 1.5 $\mu$  diam. slender, with wall encased in crystalline material; penicillus 25 to 40, or even 75 $\mu$  long, smooth, or ? *encrusted* or *sheathed* with crystalline material, figured as a main axis and terminal verticil of sterigmata or with 1 or more branches or metulae from the next node; either short or much longer than the continuation of the main axis; metulae 24 to 30 or rarely 60 $\mu$  by 1.8 to 2.5 $\mu$ ; sterigmata 7.5 to 11 by 2.8 to 3.5 $\mu$ , 1, 2 or rarely 3 together; conidia oblong (?) subglobose 4.5 by 3.5 to 4.5 $\mu$ , few or scantily produced;

Biourge no. 180 (not received) was obtained from Amsterdam (Westerdijk) but was doubtfully placed as *P. lagerheimi*. Biourge was hardly justified in proposing a Latin diagnosis for a species whose identity is so uncertain as this culture seems to have been and whose description differs so greatly from that of the type.

428. *P. desciscens* Oudemans. Arch. Neerlandaises des Sc. Exactes et Nat. 1902: 288 (289?), Tab. XXIV, fig. 1-5. See also Oudemans in Nederl. Kruiddk. Arch. Ser. 3. 2: 907. 1903. See Westling Arkiv för Botanik 11: 147. 1911.

Colonies on soil-extract gelatine, yellow green (?); conidiophores figured as arising from prostrate or submerged hyphae, septate, straight, with occasional divergent penicillus-bearing branches distant from the terminal penicillus; penicillus a single verticil of 4 metulae 9 to 12 $\mu$  long

each bearing 4 more or less divergent sterigmata about  $10\mu$  long, with loosely parallel chains of conidia; conidia described as 2 to  $3\mu$  in diameter, but figured very definitely as elliptical as noted by Westling.

Species described by Oudemans found in forest humus in Holland by Koning, listed by Waksman Soil Science 1. (Is there any fungus flora of the soil?), p. 576, 1917, and Soil Fungi and their activities, Soil Science 2: 132, fig. 10, 11, 1916; by Jensen Cornell Agr. Exp. Sta. Bul. 315, p. 485, fig. 121, 1912, whose culture was green-gray to blue-green, more or less zonate in age; reverse yellow green to red when sugar was present, with conidiophores 80 to  $500\mu$  long, with conidia ellipsoidal to globose  $2.5$  to  $3.5\mu$  in long axis.

Occasional cultures with general morphology suggesting *P. desciscens* and *P. humicola* are obtained in miscellaneous work, especially where soil contaminations are frequent. These forms lack the intensity of green in the conidial areas, and the abundant yellow and red colors in reverse and substratum so common in the biverticillate series. They are characterized by loosely fibrous gray green or bluish green surface growth, by long divergent chains of elliptical conidia, and the typically biverticillate penicillus with few metulae and few sterigmata to the verticil.

The following cultures would fall in this group no. 4115.2 from moldy paper; no. 4277 from Washington; no. 4078 D 33 from Waksman in New Jersey; no. H 5118.6 from Madison, Wisconsin, on wood; no. 4090.2 AB from Pullman Washington; no. 4157.675 from Idaho.

429. *P. exiguum* Bainier. Bul. Soc. Mycol. France 23: 96, Pl. X, fig. 5. 1907.

Colonies on licorice sticks, floccose up to 6 or 8 mm. in depth, white, tardily reddish gray brown; conidiophores short, commonly less than  $50\mu$  (13 to  $31\mu$  reported) by  $2.8\mu$ , figured as diverging branches and terminal segments of trailing or ascending hyphae; penicillus a single verticil of sterigmata, or a symmetrical biverticillate group of 3 to 5 metulae bearing sterigmata or a 1-sided verticil of 1-several metulae; metulae described only as short; sterigmata about  $8\mu$  long figured as tapering gradually to the conidial apex; conidia cylindrical-elliptical 4 by  $2\mu$ , reddish-brown, figured in parallel chains.

Species found upon a Brie cheese in France. Bainier's figure gives the aspect of the biverticillate species so definitely as to establish a presumption of relationship to that series. It was noted by Bainier as difficult to isolate and to maintain in culture. This species has not

been surely identified by others. Study of Bainier's figures shows that he failed to associate the general structure of the biverticillate penicillus with a series of related organisms hence in making his figures more or less diagrammatic, the failure of his interpretations introduces a vagueness in his drawing of members of this group.

430. *P. hirsutum* Bainier and Sartory. Bul. Soc. Mycol. France 29: 373-376. 1913.

Colonies upon licorice sticks with conidial areas blue green (C.d.C. 398, 399, 397, 393); conidiophores 230 to 780 by 3 to 4 $\mu$ , bearing a terminal verticil of metulae and occasionally at lower nodes of the stalk one or more long divergent branches with separate biverticillate penicilli; metulae 20 to 28 $\mu$  in length usually 2 to 4 in the verticil; sterigmata 15 to 22 $\mu$  long and 3 to 5 in the verticil; conidia globose 2.5 to 3 $\mu$ , fairly uniform, in long loosely parallel or diverging chains.

Sclerotia produced in some media; perithecia not found.

Optimum temperature 35° to 37°C., minimum 20°; no growth above 50°C. Cultures grew well in all common media, but did not coagulate milk, did not decompose urea, produced no action upon starch, fermented glucose, did not liquefy gelatine, produced no color in potato; consumed cane sugar without inverting it. It was not pathogenic to laboratory animals.

Except for the occasional production of sclerotia recorded by Bainier and Sartory, this species appears to be closely similar to Oudemans' *P. desciscens*, or perhaps to *P. Rolfsii* (no. 426). Some such organism may have been the basis of Hanzawa's report of *P. canum* Preuss.

## CHAPTER XXI

### THE POLYVERTICILLATE-SYMMETRICA

Division IV. *Polyverticillata-symmetrica*. Part of Costantin's Synpenicillium but excluding the type species *S. album*.

Colonies so far as known producing trailing aerial hyphae and ropes of hyphae, (also never green?); bearing conidiophores as short septate branches, with symmetrically polyverticillate penicilli producing compact masses; conidia elliptical, not in slimy masses.

The polyverticillate and symmetrical species form a group of uncertain relationship; morphologically they come between the biverticillate

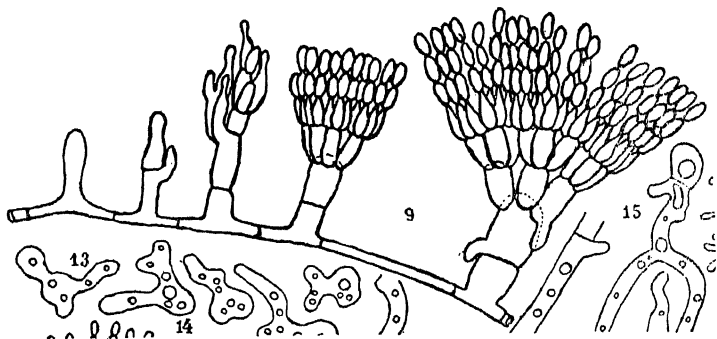


FIG. 87. *P. albicans*: Bainier's figure showing the development of the symmetrically polyverticillate penicillus.

series on the one side and Gliocladium and Scopulariopsis on the other. Biourge put them in his section *Anomala* along with the *P. brevicaulis* or *Scopulariopsis* series of species with which he reported them as sharing the biochemical character of emitting arsene when arsenic in any form is present in the substratum. Bainier regarded them as a section or subgenus in *Penicillium* to which he assigned the name *Synpenicillium* which was given by Costantin as a generic name for the species discussed by Bainier as *P. costantini*. Thaxter apparently from the literature regarded some of them as species of *Gliocladium*. The lines of separation are perhaps imaginary but certain of those species are described as producing conidia in chains without mentioning a slime-ball phase.

We have seen several species in culture and they comply closely

enough with the descriptions given by Bainier to justify believing that they form a fairly homogeneous series. Bainier's discussions are therefore reproduced in terms approximating those used for the commoner types of *Penicillia*.

One of Bainier's figures, only, is reproduced. Close scrutiny shows that his figures are so far idealized as to lose much of their definiteness of relation to particular material. Figure 87 presents the type of penicillus characteristic of the series in which there may be five or even more superposed verticils.

440. *P. albicans* Bainier. Bul. Soc. Mycol. France 23: 18, Pl. V, fig. 8, 9. 1907.

Conidiophores figured as short perpendicular branches from trailing hyphae, much larger in diameter than the sterile hyphae, consisting of 1 or 2 swollen cells, bearing a penicillus figured as regularly 2 to 3 verticillate with elements coarse, short, vesiculose rather than tubular successively smaller in diameter, and with occasional branches directed backward from the upper cell of the conidiophore or the first verticil of branches; conidia oval, at first white then slowly fawn to reddish in age.

Species common on wet straw in France and the brief note given by Bainier says its morphology is exactly that of *P. rubescens* Bainier except that the conidia are oval and a little smaller.

We have not seen this species.

441. *P. insigne* Bainier. Bul. Soc. Mycol. France 22: 134; Pl. IX, fig. 5-12. 1906.

Colonies on licorice sticks broadly effused, as a network of interlacing ropes of hyphae from which the conidiophores arise as verticils of branches; conidiophores arising as branches perpendicular to cells in prostrate hyphae or ropes of hyphae, much larger in diameter than the vegetative cells connecting with them, dividing into a basal cell which frequently produces several rhizoid-like branches, and an upper cell with rarely a second cell branching at right angles, which develops into the stalk and penicillus, with the stalk figured as coarse, unseptate very short or up to  $280\mu$  long by about  $11\mu$  in diameter producing the penicillus at its apex as a verticil of (not over 7) branches, bearing metulae, sterigmata and chains of conidia; branches up to 11 by  $5.6\mu$ ; metulae about  $6.8$  by  $4.2\mu$ ; sterigmata  $8.4$  to  $11.2$  narrowing gradually to produce the conidia which are elliptical  $5.6$  by  $2.8\mu$ , when ripe.

*P. insigne* Bainier, "*Licipenicillium*" *insigne* Brefeld and Lalliputia

*Gaillardii* Boud. and Pat. are all regarded by Thaxter as names for *Gliocladium penicilloides* Corda. Bainier's detailed description points to the conclusion that he had a form different from the common *Gliocladium* as suggested by Thaxter.

No species resembling this has been seen by us. Unfortunately there are so many items lacking in Bainier's description as to make it difficult to place his species accurately.

Recapitulation of Bainier's notes may be justified: Colonies cultivated on sticks of licorice, show conidiophores supported by a group of digitate branches at base; very slow development—during winter, requiring more than a month; aerial mycelium broadly effused, first arising as ropes of interlacing filaments which give rise here and there perpendicularly to the root-like branches and to the fruiting apparatus, at first in the form of an oval cell which quickly divides by a cross wall into two parts; the lower of these cells becomes cut off by walls from the filament bearing it. This cell is then strictly marked off by these walls into a constant and characteristic basal cell. In the first fructification, this basal cell simply increases in size, retaining more or less fully its primitive form and rarely giving rise to mycelial filaments; as the plant becomes older and more vigorous especially if the culture is growing or on agaric on other loose substratum this basal cell becomes considerably modified, swells and gives rise to five or six long digitate branches, sort of suckers or grappling hooks, which spread out more or less.

The upper part of the oval cell forms the supporting cell of the penicillus. In the earlier fructification this support (conidiophore) is rudimentary, forming a sort of "bonnet a peine"—fool's cap—a little larger than the basal cell, but considerably elongated and becoming in the normal fructification a long cylindrical cell up to 280 by  $11\mu$ , unseptate, usually straight, but sometimes sinuous or contorted.

The wall separating this cylinder from the basal cell remains often plane but at times splits at the edges with the bulging of the cells so that the surfaces in contact are convex. The far extremity of the supporting cell is always rounded (like a skull-cap) and upon it are arranged a crown of fruiting branches which are very characteristic. The number of these branches is irregular (not more than 7). Each of these is very simple. A large cell bears at its apex 3 to six smaller cells each of which bears 3 to 6 sterigmata. The large cells are variable in form—oval 11 by 5.6. As a rule in fully developed cases many of them are much smaller. The cells bearing the sterigmata are relatively oval about 6.8 by 4.2 sterigmata are gradually narrowed to the apex, vary in

length 8.4 to 11.2, and bear chains of conidia. These conidia have sometimes an oval form, sometimes oblong (?) commonly 2.8 by 5.6 or much smaller in younger fructifications.

Sometimes the basal cell bears a second conidiophore.

*P. niveum* Bainier. Bul. Soc. Mycol. France 22: 134, Pl. IX, fig. 1-4. 1906.

Not *P. niveum* Sopp.

Colonies on licorice sticks white, with surface growth consisting of anastomosing hyphae and ropes of hyphae bearing the conidiophores as perpendicular branches; conidiophore up to 650 by 12.5 $\mu$ , septate; penicillus symmetrically 5 to 7 times verticillate, branches in the primary verticil much longer than those in the secondary and later groups; sterigmata in fours or fives; conidia cylindrical to elliptical 8.4 to 11.2 by 2.8 to 3 $\mu$ .

We have not identified this organism.

443. *P. rubescens* Bainier. Bul. Soc. Mycol. France 22: 207, Pl. XI, fig. 7-13. 1906.

Colonies on licorice sticks, at first white and consisting of trailing ascending and interwoven hyphae and ropes of hyphae making a mass up to 5 mm. deep, becoming a reddish or rusty color with the development of mature conidial areas; conidiophores arising as short perpendicular branches from the aerial mycelium, figured as consisting of 1 to 3 short, heavy cells supporting a penicillus consisting of 3 to 6 times verticillate branches, more or less symmetrically produced, successively shorter and smaller in diameter, with occasional reversed branches in the primary series growing back toward the basal hyphae as supporting (?) cells; conidia elliptical 2.8 to 5.6 $\mu$  in long axis becoming rose then brownish red in ripening and giving the color to the colony.

Culture: Bainier. Our no. 4136Q43 among Waksman's cultures from New Jersey soil repeated this morphology closely enough for a provisional identification.



## CHAPTER XXII

### GLIOCLADIUM

*Gliocladium* of Corda as interpreted in descriptive literature includes species whose conidial apparatus is so *Penicillium*-like that they could readily be assigned to *Penicillium* as well as species which diverge in morphology sufficiently to justify generic segregation. In those studied by us, colony habit and appearance diverge fairly widely from the usual *Penicillia*. Nevertheless when someone takes time to straighten out the relations between these genera and reexamines all the forms described as *Gliocladium* we may explain such riddles as *P. repens* of Cook and Ellis which Ellis found abundantly about 1880 upon dead twigs of Newfield, New Jersey, and no one has since recognized (see no. 670). No consistent effort to cover all of the *Gliocladium* literature has been made but enough will be presented to exhibit the contrasts between the hyphomycetes of these two genera and to consider certain species, some of which have been described and more or less commonly regarded as *Penicillia* and others of which show suggestive relationship.

Corda's discussion of *Gliocladium* may be considered first by repeating the generic discussion already given in Chapter IV.

*Gliocladium* Corda, *Icones Fungorum* IV: 30-31, Taf. VII, fig. 92. 1840. Type species *G. penicilloides* Corda, *Icones Fungorum* IV: 30-31, Taf. VII, fig. 92. 1840.

Latin description repeated in *Icones* V, p. 14, 1842, with a brief paragraph in German. See also Matruchot. *Rev. Gen. Bot.* 7: 321, Pl. 16, 1895, also Bainier, *Bul. Soc. Mycol. France* 23: 111-112, Pl. XV, 1907.

The essential characters given by Corda were: Conidiophores erect septate, penicillately branching above, branches and branchlets septate, appressed, forming a solitary gelatinous head; conidia unicellular borne upon the tips of branchlets and held together by mucilaginous substance in a dense mass.

*Gliocladium* was thus described as reproducing the growth habits, mycelium, conidiophores and conidial apparatus of *Penicillium* except that the conidia borne successively from the tips of sterigmata become enveloped in mucilaginous drops which increase in size with the in-

creased numbers of conidia. The masses upon adjacent sterigmata fuse, then fuse with those from adjacent penicilli often forming large balls of conidia.

Matruchot has described perithecia and ascospore formation in certain species, but the forms constantly encountered in culture are purely conidial. Comparative studies of structure in both conidial and ascosporic forms are necessary before *Gliocladium* and *Penicillium* or its ascosporic sections can be safely placed with reference to each other among the Ascomycetes.

Corda in his *Prachtflora* also described as *Clonostachys*<sup>1</sup> *araucaria* a penicillate organism which he figures as producing columns of elliptical conidia in which the long axis of the conidium stood diagonally across the axis of the column so that the conidia forming the column were adherent side by side instead of end to end as in *Penicillium* or being enveloped in slime as in *Gliocladium*.

In the description of *Gliocladium*, the conidia enveloped in balls of slime have formed the character most emphasized. The conidial apparatus as observed and figured is superficially *Penicillium*-like especially in material washed in alcohol and mounted for examination, hence species have been placed sometimes in one genus and again in the other. Few attempts to establish real relationships have been recorded. From the standpoint of relationship with *Penicillium* the process of conidium formation becomes significant.

The conidium producing apparatus as seen and as figured in *Gliocladium* is penicillate. The typical cell of the group is the sterigma which cuts off conidia from its apical tube. The primary branching system varies from asymmetrical to symmetrical. Most of the described species approach the symmetrically branching system of the *Polyverticillata*. The diameter of the main conidiophore is usually much greater than that of the primary branches and the elements in each of the successive stages of branching are smaller. The sterigmata vary from shades and measurements of those in typical *Penicillia* to long subulate tubes. In some species as also in the *Polyverticillata* a reflexed

<sup>1</sup> See Chapter IV.

*Clonostachys* Corda, *Prachtflora*, p. 31, Tafel XV. Type species: 450 *C. araucaria* Corda.

Mycelium creeping, continuous (?); stalks erect, simple, continuous, verticillately branched above; each branch bearing 2 or more superposed verticils of 4 sterigmata each at successive nodes; sterigmata (ramuli) subulate with apex subcapitate bearing spores spirally forming a kind of spike; spores unicellular, with walls hyaline and contents curved around a central globule.

series of branches toward the base of the conidiophore turn back to form a supporting claw like base attached to a rope of mycelium or actually reëntering the substratum.

In *Clonostachys*, Corda described conidia as produced upon a subcapitate apex in a special manner but his figure shows no details and no evidence that he actually saw such a disk. In certain strains of the rosy section of this group many penicilli showing the arrangement of conidia seen in Corda's figure have been studied and give us every reason to believe that approximately Corda's organism was before us, even though the numbers of elements in the verticil were not fixed at four,

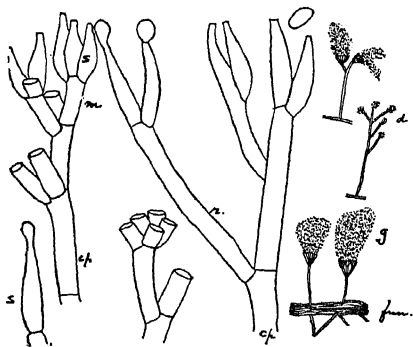


FIG. 88

FIG. 88. *G. roseum* type of penicillus: Thom's 1910 figure 15, showing the more *Penicillium*-like conidial masses in various stages of formation; *ch*, conidiophore; *r*, branch; *s*, sterigmata; *g*, gelatinous mass of conidia; *fun*, rope of hypha; *d*, divaricate branches with spore balls.

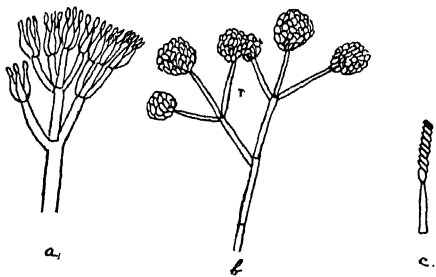


FIG. 89

FIG. 89. Diagrammatic sketches of structures which can sometimes be found in some preparation: *a*, the *Penicillium*-like branching system; *b*, the *Aerostalagmus*-type; *c*, the sterigma of the *Clonostachys* type as interpreted here.

as Corda believed. The colonies studied and the conidiophores and penicilli examined in detail were not different in general structure from certain species of *Gliocladium* and from the type of organism sometimes reported as *P. roseum*. (Thom 1910). Conidium production in the three forms may therefore be studied together.

Microscopic examination shows that the conidia are cut off successively from the tips of sterigmata as in *Penicillium*. In studying various areas and different ages of petri dish colonies of certain species some areas are found in which the conidia remain for a time in chains.

In other areas of the same colony, penicilli show the *Clonostachys* types of conidial columns and in still other areas, the conidia are found massed in the typical slime balls.

Correlation of these observations with cultural conditions, shows the *Penicillium*-like arrangement of conidia as primitive and regularly occurring when careful search reveals penicilli in the process of formation. One species, *P. vermoeseni* Biourge with the colony characters of a *Gliocladium* produces separate chains of conidia persistent as in *Penicillium*. In another species, *G. catenulatum* Gilman and Abbott, the conidia remain end to end in chains and the chains become adherent into a column perhaps a little more firmly bound than in many of the true *Penicillia* but differing little in superficial appearance as we have studied both. In another species the *Penicillium*-like penicillus with conidia all remaining in chains may be found to persist in penicilli developed late in the growth period when drying of the substratum has nearly reached the point at which growth ceases. Penicilli, in the area produced during the rapidly growing period have all become slime balls; in intermediate areas whole groups of penicilli show the *Clonostachys* type of conidial mass.

The hypothesis offered in explanation is that in a fairly homogeneous group, we have such a species as *P. vermoeseni* Biourge in which the chains of conidia persist and remain separate as in *Penicillium*; the *G. catenulatum* type with chains massed into columns; the *Clonostachys* type of organism with the outer conidial wall partially deliquescent and the elliptical spores slipping back in a direction determined by the particular shape of the conidia, until sufficient surfaces are in contact for the viscous slime present to support the weight of the conidium or even the whole column of conidia. The extent of the deliquescence thus determines whether conidia in several chains will remain there as produced, or will adhere into columns as figured by Gilman and Abbott for *Gliocladium catenulatum*, or every conidium becoming loose from its fellow at the point of contact will slip back past its fellow toward the base of the chain so that all assume the position diagonal to the axis of the column, as in *Clonostachys* or all slip back to form a composite slimy globule of conidia and slime as in the typical species of *Gliocladium*.

From such a series of observations, the generic name *Clonostachys* is seen to rest on very slight foundation, if any, and that probably *Clonostachys araucaria* Corda should be regarded as a member of the rosy series in *Gliocladium* with the enlarged tip regarded by Corda as a conidium-producing vesicle actually forming the newest conidium just developing on the tip of the sterigma.

Three series of more or less contrasting members of the aggregate *Gliocladium* are commonly encountered in culture.

(1) *G. roseum* series. The rosy or salmon series shows a fairly complete gradation from forms near to *P. vermoeseni* with its penicilli like a true *Penicillium* through *Clonostachys araucaria* with the end to end relations of conidia broken and each cell slipped half its length backward to the *G. (or P.) roseum* form in which the slime balls have become well established.

(2) *G. deliquescens* series. A gray-green to fuscous series, represented by such forms as *G. deliquescens* Sopp fide Gilman and Abbott, common in the soil and showing widely spreading submerged mycelium with areas or clumps of conidiophores and penicilli above the surface. The conidia of these forms as far as known to us are found regularly in slime balls.

(3) *The floccose-green series*. A bright green and floccose series in which the colonies show abundant radiating floccose or funiculose aerial hyphae with abundant green masses of penicilli distributed in characteristic manner species by species. Here again we have a transition species in no. 451 *G. catenulatum* Gilman and Abbott as already indicated in which the conidia remain in chains which adhere into columns.

*Transition species*. As indicated already the transition from the penicillus of the true *Penicillia* toward *Gliocladium* is illustrated by two described organisms, *P. vermoeseni* in the white to pink or salmon series, and *G. catenulatum* Gilman and Abbott in the green series. These two descriptions are introduced here.

451. *G. vermoeseni* (Biourge) Thom. *P. vermoeseni* Biourge. Monogr. La Cellule 33: fasc. 1, p. 230; Pl. XXIII, fig. 137. 1923.

Colonies on wort gelatine, producing numerous salmon colored coremia 10 mm. in height or more; conidiophores about  $5\mu$  in diameter; metulae 7 to 15 by 2.5 to  $5\mu$ , irregularly borne, irregular in number or none; sterigmata 10 to 20 by 2.5 to 3.5, in groups of 2 to 5, or even 7; conidia elliptical 5 to 7.5 by 3 to  $4\mu$ .

Habitat: In certain species of *Areca*, parasitic or semi-parasitic.

Biourge calls his no. 415 which was not sent to us a *Stysanus* with \* flesh-rose or salmon color in nature and in culture. Later culture no. 4876.15 was received from Dr. Westerdijk under this name and noted as having two forms of conidia, a rosy form and a green form. The two forms were readily separated and the rosy form was compared with Biourge's discussion of *P. vermoeseni*. There is good reason to believe this to be Biourge's organism and probably his type strain transmitted

through several workers. The following description is based upon our own cultures:

Colonies on Czapek's solution agar, loosely floccose, broadly spreading covering the entire surface of the substratum in petri dishes and following the glass to the edge and reflected back on the inside of the cover, or even out under the edge of the cover, surface unevenly, loosely, massed, white becoming tardily salmon with the development of ripe conidial masses; hyphae sinuous, colorless, coarse, 3 to  $6\mu$  in diameter, showing large and numerous vacuoles; reverse colorless, then yellowish to greenish yellow; odor evident, peculiar; conidiophores mostly as short branches of trailing interlacing aerial hyphae and ropes of hyphae about 100 to 200 by 4 to  $5\mu$ ; conidial apparatus produced, variously, partly as single sterigmata produced as terminal cells on short branches, or as variously branching systems, at times truly penicillate, and forming irregularly distributed masses white then salmon or rosy especially on the glass and at the junction of top and base of petri dish; metulae when recognizable 10 to  $12\mu$  by  $3\mu$  at base and with much enlarged apex; sterigmata very irregular in size, 8 to 10 to  $12\mu$  or up to  $20\mu$  or more when forming a whole branch; conidia 4 to 6 by 3 to  $4\mu$  elliptical, colorless, somewhat irregular at first, elliptical when ripe, and forming chains, 1 to 2 mm. in length in old cultures and adhering in large masses which break off.

The conidial apparatus found here allies this species closer to the *Gliocladium* than to *Penicillium* hence its allocation here.

*Gliocladium catenulatum*. Gilman and Abbott. Iowa State College Jour. Sci. 1(3): p. 303, fig. 37. 1927.

Colonies on Czapek's agar pure white, spreading, floccose, becoming olive green to bright green in the center as fruiting areas develop, and clear dark green in old cultures; fruiting areas are usually confined to center of colony and one or two concentric zones separated by sterile mycelium; reverse colorless to yellowish. Aerial mycelium abundant, simple or in ropes, from which the conidiophores arise as branches. Conidiophores often once and sometimes twice branched, coarse, pitted or rough, 50 to  $125\mu$  long. Heads are composed of conidial chains in long, close columns, enveloped in slime, up to  $150\mu$  long. Fructification in three stages, elements of fructification pitted or rough; primary branches 15 to  $20\mu$  by 3.5 to  $4\mu$ ; metulae 7 to 9 by 15 to  $25\mu$ ; phialides 10 to  $20\mu$  long. Conidia elliptical, smooth, pale green, 4 to  $7.5\mu$  by 3 to  $4\mu$ .

From soil: United States: Utah.

*The G. roseum series.*

453. The Type Species: *Gliocladium penicilloides* was described by Corda in *Icones Fungorum* IV: 31. Taf. VII. fig. 88, 89. 1840.

Corda described mold colonies found fruiting upon the hymenial surface of species of rotting *Thelephora* with the following characters: Colonies small, white; conidiophores erect, flexuous, enlarging above, septate, pulverulent, colorless; penicillus with primary branching opposite, branchlets verticillate in fours, appressed; heads of conidia globose, white; conidia  $6\mu$  in long axis, oblong, embedded in a mucilaginous mass.

Matruchot described ascospore formation in *G. penicilloides* Corda. Winter in *Rabenhorst Krypt. Fl.*, 2 Aufl., I abt., 2, p. 61, 1887, suggests that *Eurotium insigne* Winter is probably the ascosporic form of this species.

By this description as pointed out by Gilman and Abbott, the type of the genus falls in the non-green section—designated white (by Corda) for the type species. As seen in culturing many of them white pass slowly into cream and many gradations from white to rosy, pink, or salmon shades can be found. Representatives of this rosy to salmon series have found their way into many collections. One of them was included by Thom 1910, pp. 49, 50, fig. 15, as *P. roseum* Link. It was purchased from Kral in Prague. This description and the notes as to its distribution, with the substitution of *Gliocladium* for *Penicillium* as first suggested by Bainier follows:

454. *G. roseum* (Link?) Bainier. *Bul. Soc. Mycol. France* 23: 111–112, Pl. XV, fig. 1–6. 1907.

Synonym *P. roseum* Link. *Obs.* II, p. 37, 1816; see also Link, *Sp. Plant* Ed. 4, Vol. 6, pt. 1, p. 69, 1824; Fries *Sys. Myk.* 3, p. 409, 1829; *Pers. Myc. Europ.* 1822.

Compare Thom, *U. S. Dept. Agr. Bur. Anim. Ind. Bul.* 118, p. 49, fig. 15, 1910. This is apparently identical with DeThümen, *Myc. Universalis* no. 1179 collected in South Carolina by Ravenel. Whether the organism commonly cultivated as *P. roseum* is the one described originally may be doubtful but the wide use of the name without confusion appears to justify the identification. Under strict interpretation of generic definitions we have seen single cultures of this organism with fruiting branches determinable as *Penicillium*, *Gliocladium*, and when old, *Clonostachys*. Bainier's type, our no. 4640.428 confirms the belief that these strains belong together. Thom's description follows:

Colonies on milk-sugar gelatin or potato agar white to pink or salmon

in fruiting areas, loose floccose with simple hyphae and ropes of hyphae, producing dense irregular pinkish masses up to 1 mm. or more deep in old cultures; conidiophores bornes as perpendicular branches of aerial hyphae or ropes of hyphae 45 to 125 $\mu$ ; penicilli up to 140 $\mu$  in length, once or twice irregularly alternately or verticillately branched, with sterigmata varying from 12 by 2 to 3 $\mu$  in verticils of 5 or less to 17 by 2.3 $\mu$  when solitary, bearing conidia which become aggregated into gelatinous balls or masses; conidia colorless (pink or rosy in mass), elliptical, 5 to 7 by 3 to 5 $\mu$ , slightly apiculate, smooth, appearing delicately granular within; colonies liquefy gelatin cultures rapidly and give an alkaline reaction to litmus media.

Bought from Kral, in Prague, Bohemia. Compare figures 88 and 89.

Closely similar organisms have been found occasionally in this laboratory; received once from a correspondent in Halle, Germany, and later found under this name as no. 1179 in DeThümen's Mycotheca Universalis as collected by Ravenel in South Carolina in 1876 upon leaves of *Buxus*; this and several other specimens were found in the mycological collection of the Bureau of Plant Industry, United States Department of Agriculture. The spores are the same length as given by Saccardo, but slightly broader. The number of specimens found under this name from widely different workers appears to justify the belief that this is the organism described by Link under this name. If the development of a mucilaginous mass enveloping the conidia be regarded as a sufficient basis for separation of such species under the generic name of *Gliocladium*, this species would become *Gliocladium roseum* (Link). It is cited so and the change attributed to Thom by Gilman and Abbott.

The form upon *Buxus* is cited by Saccardo, referring to it as "*P. roseum* Cooke, non-Link," and held to be *Verticillium buxi* Auersw. et Fleisch.

*P. roseum* is cited by Berkeley and Broome as on *Hibiscus* in Fungi of Ceylon, no. 911, Jour. Linn. Soc. Bot. 14, p. 101, 1875.

*P. roseum* var. *coremioides* Kickx Fl. Fland. II, p. 306 appears to be another strain of the series.

Bainier uses the name as a new species for what appears to be one of the series in Bul. Soc. Mycol. France 23: 111, Pl. XV, fig. 1-6.

Bainier in figuring his strain showed large columns of conidia about halfway between *Clonostachys* and *Penicillium*, and the typical slime balls upon divaricate sterigmata characteristic of the young stages of the colony. Although we placed a question mark as to the identifica-



tion of Bainier's material by Thom's description, we have his culture and we have put these together under this description rather as representing a series of widely distributed strains than a definite clear cut designation of particular organisms. No attempt to differentiate them will be made here.

455. *Acrostalagmus roseus* Bainier, Bul. Soc. Mycol. France 21: 225-227, Pl. 12, fig. 1 to 9. 1905.

Colonies upon licorice sticks, at first white, septate, creeping and spreading broadly and hyphae combined into prostrate fascicles or ropes, bearing branches 60 to 80 $\mu$  by 2 to 4 $\mu$ ; penicillus figured and described as with or without 1 or more primary branches, then a secondary series or verticil of metulae, bearing sterigmata 3 to 5 in the verticil and 16 by 2 $\mu$ , bearing the conidia which form a mucilaginous ball extending across the tips of the whole group of sterigmata, and a second form of penicillus with the branches more divergent, several times verticillate producing a hemispherical mass of branches with sterigmata over the whole surface and ultimately becoming more or less completely covered by the spores imbedded in a continuous mass of mucus; conidia ovoid or occasionally globose, 2 to 6 by 2 to 3 $\mu$ , and remain viable for a year.

Species found upon seeds of millet which birds dropped on moist soil as rosy hemispherical masses on the surface of the grain.

456. *Isaria clonostachoides* Pritchard and Porte. Phytopathology 12: 167-172, fig. 1 and Pl. XII. 1922.

On artificial media and tomato fruits bundles of agglutinated hyphae forming a cylindrical mass, splitting into free filaments toward the apex, 2 to 5 mm. high, septate, bearing elongated conical fruiting masses each composed of repeated whorls of 2 to 4 branches or subulate branchlets; conidia on free hyphae or conidiophores, hyaline to pink or pale salmon in globular and spike-like masses of mucus, curved, not similar at each end, 5 to 8.5 by 3.2 $\mu$ . Temperature range 7° to 36°C., optimum 29°C.

On green and ripe tomato fruits, Arlington, Va., and District of Columbia, 1919 to 1921.

II. *G. deliquescens* series. The second of the three series includes strains with the vegetative mycelium mostly submerged when seen in cultures. Such colonies have appeared in cultures made from soil as seen by us in Connecticut, in Washington and in various collections submitted for identification. The same observation has been commonly reported in the literature.

Gilman and Abbott have reported one of these (our no. 4894.17) as *P. deliquescens* Sopp. This appears regularly in our own cultures from soil. Sopp's figures appear to have been drawn from colonies grown upon the stem of some phanerogam rather than upon laboratory culture substrata although reactions upon various media are given. We may tentatively accept the identification by Abbott as at least bringing together organisms of the same series and give both descriptions as more adequately representing the range of morphology encountered. *G. viride* Matruchot appears also to belong here.

457. *G. deliquescens* Sopp. Monogr., pp. 89-93, Taf. I, fig. 1-6. 1912.

Colonies clear yellowish green becoming darker in age, at first a typical area of crowded, *Penicillium*-like conidiophores which later become enveloped in slimy masses as the conidial chains dissolve and run together; reverse gray at first, later dark green almost black, odor characteristic: gelatin liquified; conidiophores up to 1 mm. long, erect, coarse, septate, 1 to 5 times penicillate branching, each series of branchlets progressively smaller so that the sterigmata are much smaller in diameter than the primary branches; conidia about 1 by 1.5 to  $2\mu$  or somewhat larger when ripe, at first fusiform, later more rounded at the ends, at first and on different media for varying periods in chains which break up as the conidia become enveloped in masses of slime; perithecia and sclerotia not found.

Species found upon a specimen of *Daedalea unicolor* in Norway; colonies grew well upon various media and remained viable for three years.

Growth is vigorous at  $33^{\circ}$  to  $37^{\circ}\text{C}.$ , with optimum about  $35^{\circ}\text{C}.$ , continues up to about  $40^{\circ}\text{C}.$  Conidia are still found at  $+1^{\circ}\text{C}.$

Sopp's type has not been seen. Gilman and Abbott (cit. p. 304) described an organism under the name as follows with figures which we readily recognize in our own cultures.

"Growth not abundant on Czapek's agar. On bean agar, broadly spreading, producing a thin, transparent growth of sterile hyphae over the entire medium, from which the dark green fruiting areas soon develop; surface deep, dark green to blackish green; reverse colorless. Aerial mycelium scant, colony consisting almost entirely of conidiophores and slimy heads. Conidiophores arise from submerged and surface hyphae, several from one point; both aerial and submerged stolons present at these points; conidiophores 100 to  $225\mu$  by 8 to  $10\mu$ . Fructification typically in four stages, consisting of three to five

primary branches arising from the apex of the conidiophore; these bear a verticil of secondary branches, and these verticils of metulae; phialides closely crowded on the metulae, club shaped; primary and secondary branches and metulae elongate oblong, slightly inflated at the apex. Primary branches 15 to 20 $\mu$  by 3 to 3.5 $\mu$ ; secondary branches 13 to 15 $\mu$  by 3 $\mu$ ; metulae 8 to 10 $\mu$  by 1.5 to 2 $\mu$ ; phialides 6 to 8 $\mu$  by 1 to 1.5 $\mu$ . Conidia elliptical, greenish, smooth, granular within, 3 to 3.8 $\mu$  by 2 to 2.5 $\mu$ . Hyphae, conidiophores, and elements of fructification coarse and pitted, or rough. Slime production very abundant, usually enveloping the entire colony."

458. *Gliocladium atrum*, Gilman and Abbott. Iowa State College, Jour. Sci. 1(3): p. 305, fig. 40. 1927.

"Colonies on Czapek's brown green, small, slowly spreading, largely submerged; aerial mycelium olivaceous, scanty, aerial growth consisting mostly of conidiophores; colonies moist with slime which envelops the heads. On bean agar considerable aerial mycelium is produced. Conidiophores arise mostly from submerged hyphae, olivaceous, thick walled, smooth, septate, often slightly flexuous, 75 to 300 $\mu$  by 3 to 4 $\mu$ . Conidial heads enveloped in slime, round, chains not distinguishable; fructification typically in three stages, sometimes in two or four. Primary branches oblong, 8.5 to 9.5 $\mu$  by 3 to 3.5 $\mu$ ; metulae oblong, 7.5 to 9.5 by 3 $\mu$ ; phialides flask-shaped, 7.5 to 10 by 1.5 to 2.5 $\mu$ . Conidia oval to ovoid, smooth, light green to almost hyaline, 2.5 to 4 $\mu$  by 2 to 2.5 $\mu$ ."

From soil: United States: Louisiana.

There is sufficient color in the conidiophores of this fungus to place it with the Dematiaceae. However, the morphological structure is that of the genus *Gliocladium*, and it was placed in this genus because of its evident relationship to the other species included in this group.

III. *The floccose-green series*. The third series of species of *Gliocladium* common in miscellaneous culture work, has the floccose radiating aerial hyphae and ropes of hyphae seen in the *G. roseum* series but conidial areas some shade of green, many of them bright shades such as Saccardo's viridis which is used to designate one of these organisms and taken here as typifying the series. *Gliocladium catenulatum* already discussed has its affinities here. Members of the series with the characteristic conidial slime balls are often obtained in miscellaneous culture work with soil or with decomposing plant materials. Gilman and Abbott's discussion of *G. fimbriatum* is introduced as fairly typical of the cultures seen by us.

459. *Gliocladium fimbriatum*. Gilman and Abbott. Iowa State College Jour. Sci. 1(3): p. 304, fig. 38. 1927.

Colonies on Czapek's agar broadly spreading, orbicular, pure white at first, with zones of dark leaf green fruiting areas appearing near the center of the colony. Conidiophores arise from aerial hyphae, smooth, up to  $25\mu$  long; several from one point, stolon-like hyphae usually present at point of origin. Heads enveloped in round balls of slime in which chains are not distinguishable; fructification in two stages, with divergent branchlets or metulae which bear elongate flask-shaped, appressed phialides, or with conidia borne directly on a few finger-like phialides which arise irregularly from the conidiophore; in most heads one or more branchlets arise laterally from the conidiophore some distance below the main head; metulae elongate, extremely variable in size, phialides usually 10 to  $20\mu$  long, from flask-shaped to irregular elongate. Conidia elliptical or elongate, ovoid, smooth, pale green,  $6.5$  to  $9.5\mu$  by  $2.5$  to  $4\mu$ .

From soil: United States: Iowa, Louisiana.

No attempt is offered here to cover all of the described species of *Gliocladium* or even to reproduce our own notes of many isolations. They have not been studied sufficiently to offer a critical valuation of the various species proposed. The measurements of conidia taken from the information in hand and tabulated will give some idea of the range reported in this single feature of the group.

In this list the species described and cultures studied are arranged according to the measurements of conidia beginning with the largest.

Conidia 8 to 10 by 3 to $4\mu$ , white or gray.....	<i>G. nicotianae</i> Oudemans.
Conidia 7 to 11 by 2 to $2.5\mu$ .....	<i>G. macropodium</i> El Marchal.
Conidia $6.5$ to $9.5$ by $2.5$ to $4\mu$ , green....	<i>G. fimbriatum</i> G. & A.
Conidia 5 to $8.5$ by $3.2$ , salmon.....	<i>Isaria clonostachoides</i> P. & P.
Conidia 6 to 7 by $3\mu$ , green.....	<i>G. viride</i> cult. F. L. Stevens.
Conidia 5 to 7 by 3 to $5\mu$ , pink to salmon.....	<i>P. roseum</i> fide Thom.
Conidia 5 to 7 by $3\mu$ , pale ochraceous....	<i>G. luteolum</i> von Höhnelt.
Conidia 7 by $2\mu$ , white.....	<i>Clonostachys theobromae</i> Delacroix.
Conidia $6\mu$ in long axis, white (ascosporic?).....	<i>G. penicilloides</i> Corda.
Conidia 5 to 6 subglobose.....	<i>G. agaricinum</i> Cooke and Massee.
Conidia 5 to 6 by $2.8\mu$ ; rosy.....	<i>G. roseum</i> Bainier.
Conidia 4 to 7 by 3 to $4\mu$ , green.....	<i>G. catenulatum</i> G. & A.
Conidia 4 to 9, mostly $4.7$ to $6.7$ by $2.5$ to $4.7$ , hyaline.....	<i>G. flavum</i> Van Beyma.

- Conidia 4 to 6 by 3 to  $3\mu$ , salmon.....*P. vermoeseni* Biourge.  
 Conidia 5 by 3 hyaline toward fuscous...*G. compactum* Cooke and Massee.  
 Conidia 4.2 to  $6.3\mu$ , ascosporic.....*G. prolificum* Bainier.  
 Conidia 2 to 6 by 2 to  $3\mu$ .....*Acrostalagnus roseus* Bainier.  
 Conidia 4 to 5 by 2 to  $2.5\mu$ , colorless....2464d. Gruenberg.  
 Conidia 4 to 5 by 2 to  $3\mu$ , rosy.....2471 from S. Africa.  
 Conidia colorless, 4 by  $2\mu$ .....*G. africanum* Eichelbaum.  
 Conidia 3 to 3.8 by 2 to  $2.5\mu$ , green.....*G. deliquescentis* fide G. & A.  
 Conidia 3.5 by  $2\mu$ , hyaline.....10918.1 Humphrey.  
 Conidia 3.5 by  $1.8\mu$ , pale green; asco-  
     spores 3.5 to  $5\mu$ .....4136.M19.1.  
 Conidia 2.8 to 3 by 1.7 to  $2\mu$ .....*G. pulchellum* Penz and Sacc.  
     (New genus suggested Clado-  
     glum Penz. and Saccardo.)  
 Conidia 2 to 4 by 2 to  $2.5\mu$ , green.....*G. atrum* G. & A.  
 Conidia 2 to  $3\mu$ , globose, colorless, colon-  
     ies brown.....*P. repens* C. & E.  
 Conidia 2 to 2.5 by  $1.5\mu$  in white  
     masses.....*G. lignicolum* Grove.  
 Conidia 1.5 to 2 by  $1\mu$  or slightly larger;  
     yellow green.....*G. deliquescentis* Sopp.

## CHAPTER XXIII

### SCOPULARIOPSIS

479. *Scopulariopsis* Bainier (emended) Thom. Type species *P. brevicaula* Saccardo.

Synonyms: *Acaulium* Sopp; *Penicillium* sub-section VI. *Anomala* Biourge.

Colonies never green, with aerial hyphae partly at least in trailing and anastomosing ropes or fascicles (funiculose); conidiophores very short or wanting commonly borne along the funiculose hyphae; conidial apparatus *Penicillium*-like or consisting of varying aggregations of branches and sterigmata, at times reduced to single sterigmata scattered along aerial hyphae; sterigmata more or less specialized tapering gradually from a basal tubular section or even the base itself toward a conidium bearing apex, or narrowly tubular without tapering, cutting off conidia from the apex by cross walls; conidia more or less pointed at the apex and truncate at the base with a more or less thickened basal ring surrounding a basal germinal pore, with walls usually thickened and often variously marked or roughened.

The species appear as agents of decomposition after the usual green *Penicillia* have ceased to be active; that is in later stages of decay processes.

Sopp (Monogr., p. 33, 42-46, 1912) in describing *Acaulium* adds the observation of perithecia showing small but definite ostioles, that they produce an arsenical odor, that they decompose milk, cellulose, resinous wood, paper and sawdust, that they grow poorly on pure cotton, and that they grow at higher temperatures than most *Penicillia*.

Bainier, 1907, was probably right in separating as belonging to a genus other than *Penicillium*, the group of strains and species centering upon Saccardo's *P. brevicaula* whose general structure is well known but whose type strain is not known. These organisms have been investigated by Gosio, Ceni, Huss, and other workers in various lands on account of their biochemical usefulness in indicating the presence of minute traces of arsenic in the substratum by the evolution of arsene from the growing culture. Bainier isolated a series of forms with considerable divergence in spore and colony characters and described them as different species. Biourge finds in this type of organisms, the probable identification of

Corda's *P. anomalum* hence calls this lot of forms a sub-section *Anomala* in the genus *Penicillium*. No one who has studied many strains of this group in comparison with the usual types of *Penicillium* pretends to believe in a close relationship between them. Sopp 1912, with the same structural group undoubtedly in hand, founded his genus *Acaulium* and described a number of species, some of which were reported as producing perithecia with definite ostioles. Unfortunately Sopp failed to describe his species fully enough to ensure their identification by others thus far. Nevertheless, his data are complete enough to support the belief that this series of forms should be separated from *Penicillium*.

This conclusion is fortified by Loubière's description of *Scopulariopsis candida* as the conidial stage of *Nephrospora Mangini* as a new genus and species with perithecia in general agreeing with the description by Sopp whose *Acaulium albonigrescens* may well have been the same as Loubière's species.

Biourge discussing his sub-section *Anomala* reports (Monogr., pp. 214-216): The stalk is shortened; branching is without regular system; metulae are irregular or absent; sterigmata of the usual form are accompanied by widely divergent forms; chains of conidia may be arranged in the ordinary way, or as a separate chains on single cells; all species give Gosio's reaction with minute traces of arsenic; great variations are seen in the group; the type of the series is probably Corda's *P. anomalum*, but since it was not possible to fix upon the actual strain or species of Corda Biourge made the name "*anomala*" into a section name for the series called by Bainier *Scopulariopsis*. Harz put them in *Spicaria*. Oudemans put them in *Monilia*. Sopp proposed the genus *Acaulium*. Biourge added *Stysanus* to the series. Older workers, Fresenius, Rivolta, and Bonorden discussed these species as *Torula*, *Oidium* and often figured them.

Many of these forms tend to develop while submerged in liquid, or below the surface of any substratum used, although with distortions, swellings, vesiculation of the mycelial cells; aerial fruiting develops very slowly, partly on ropes of hyphae, partly on simple hyphae with considerable sterile areas. Sopp notes the extreme difficulty of the group to which Biourge agrees and adds that he is not satisfied with the disposition he has made. Sopp reports perithecia. Biourge finds all his observations of perithecia unsatisfactory. He has found members of this group as constant impurities in his other cultures.

Study of natural substrata shows species of *Scopulariopsis* to be abundant in every region surveyed. Miss Dale found them in English

soil; Saccardo, Gosio, Ceni and others have reported them in Italy; Pribram's collection coming from Vienna was full of them as replacements of other organisms; others from Asia, South Africa, and hundreds of strains from America have been seen. We have isolated them from soil, from many varieties of old cheese both imported and domestic, in which they are abundant in old and over-ripe products, especially Camembert. In the Camembert rooms the species of *Scopulariopsis* frequently are so abundant as to produce a characteristic ammoniacal odor. They are fairly common upon stored meat. In one lot of musty hams, cultures showed the mycelia of these forms present deep in the tissues although mustiness was the only discernible effect of their activity.

It is not surprising to find a series of them figured by Rivolta with incomplete descriptions leaving them recognizable only to the genus, as follows: *Torula rufescens* Fres. on p. 438, Tar. V, fig. 124; *T. rubiginosa* Rivolta, p. 438, Tav. V, fig. 126; *T. alba* vel. *umbilicata* Riv., p. 439, Tav. V, fig. 128; and *Oidium penicilloides* Rivolta p. 448, Tav. V, fig. 137.

This same type of observation was probably the basis of *P. toruloides* Preuss, 1852. Another was distributed in various collections as *P. onychomycosis* n. n.

480. *Scopulariopsis brevicaulis* (Sacc.) Bainier. Bul. Soc. Mycol. France 23: 99-103; Pl. XI, fig. 1-6. 1907.

Synonym: *P. brevicaulis* Saccardo in F. ital. t. 893, and *Michelia* II, p. 547.

Colonies upon licorice sticks, producing abundant mycelium, white then slowly pale rosy (avellaneous?) with the development of conidia; conidial apparatus irregular upon very short stalks (conidiophores) developing a complex penicillus with branches in several superposed series, and usually 3 to 4 in the verticil, or frequently reduced to single sterigmata or groups of sterigmata sessile or nearly so on the hyphae; sterigmata long, tapering gradually to the conidia bearing apex; conidia given as variable usually about  $6\mu$  and almost spherical, but Bainier notes also forms pointed at the apex and truncate at the base.

This species as described by Bainier was evidently the type species of the genus *Scopulariopsis* whether correctly or incorrectly representing Saccardo's species. It is therefore placed in close association with the generic discussion.

In the following list the species included are arranged into (1) questionable series, (2) perithecial and sclerotial series, then (3) into groups based upon the markings, color and size of the conidia.



## 1. Probable members of the group but without adequate description

- P. cinnabarinum* Fuckel.  
*P. brevipes* Corda.  
*P. onychomycosis*.  
*S. venerei* Greco.  
*S. arnoldi* (Mang. & Pat.) Vuillemin.

## 2. Perithecia or sclerotia reported

- A. insectivorum* Sopp.  
*S. blochii* Matruchot.  
*A. albo-nigrescens* Sopp.  
*A. nigrum* Sopp.  
*S. cinera* E-W & G.  
*P. brevicaulis* var. *glabrum* Thom.  
*Oo. glabra* Hanzawa.  
*A. flavum* Sopp.

## 3. Without such structures

## (a) Conidia described as smooth:

- 13 to 15 $\mu$ , globose.....*P. epigaeum* B. & C.  
violet brown, 12 to 15 by 9 to 10 $\mu$ .....*A. violaceum* Sopp.  
white, 12 to 15 by 8.5 to 10 $\mu$ .....*M. acremonium* Delacroix.  
amber, 12 to 13 $\mu$ .....*P. coffeicolor* B. & Br.  
yellowish, 10 to 14 by 5 $\mu$ .....*A. fulvum* Sopp.  
red, 8.4 to 11.2 by 5.5 $\mu$ .....*S. rubellus* Bainier.  
white to cream, 5.6 to 11 by 3.6 $\mu$ .....*S. communis* Bainier.  
white, 8 to 10 $\mu$ .....*S. candelabrum* Loubière.  
Pale rosy, 7 by 8 $\mu$ .....*P. benizianum* Saccardo.  
yellow, fulvous, 5 to 7 $\mu$ .....*S. rufulus* Bainier.  
avellaneous, 6 to 8 $\mu$ .....*M. Koningi* Oudemans.  
white, 6 to 8 by 3.5 to 6.5 $\mu$ .....*P. constantini* Bainier.  
rosy, 3 to 6 by 2.5 to 5 $\mu$ .....*Oo. rosatum* Biourge.  
white, 6 to 7 by 3 $\mu$ .....*P. albicans* Bainier.  
5 to 6 by 3.5 to 4.5 $\mu$ .....*P. auridorsum* Biourge.  
3 to 4 by 1.5 to 2 $\mu$ .....*S. blochii* Matruchot.

(b) Conidia described as smooth at first, tardily rough: yellowish, 6 to 7 by 4.5 $\mu$ .....

*M. penicillioides* Delacroix.

## (c) Conidia described as rough:

- white or cream, 9 to 10 $\mu$ .....*P. brevicaulis* var. *album* Thom.  
brownish, 10 to 11 by 8 $\mu$ .....*A. anomalum* Sopp.  
carneolo-isabelline, 6 to 9 by 8 $\mu$ .....*P. coccophilum* Saccardo.  
pale avellaneous, 5.6 to 8.4 $\mu$ .....*S. repens* Bainier.  
pale yellow, 6 to 7 $\mu$ .....*S. casei* Loubière.  
7 by 6 $\mu$ .....*P. brevicaulis* Saccardo.  
reddish brown, 5 to 7 $\mu$ .....*P. sacculum* Dale.  
white to pale salmon, 5 $\mu$ .....*Oo. cretacea* Biourge.

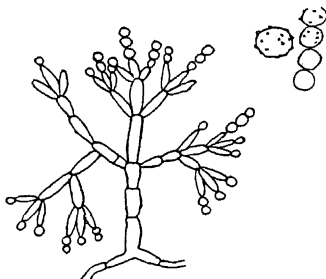


FIG. 90. *P. brevicaulis* Saccardo's original figure no. 893 in *Fungi italici*, which is obviously diagrammatic.

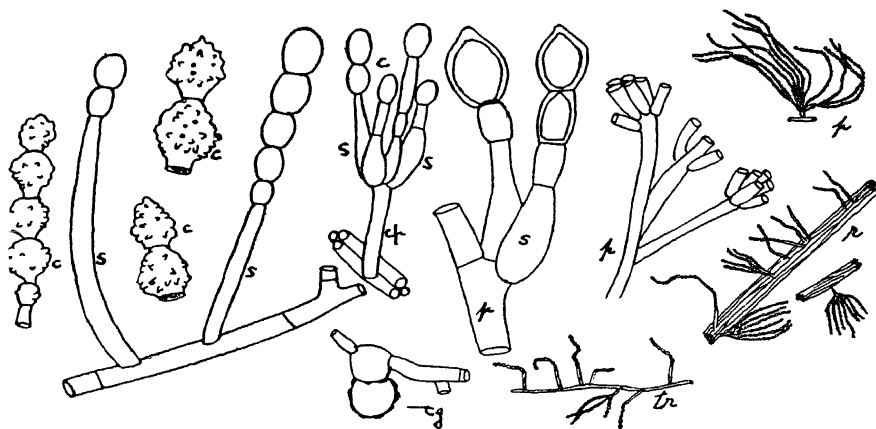


FIG. 91. *S. brevicaulis* Sacc.: Figure from Thom, 1910, showing detail of penicilli.

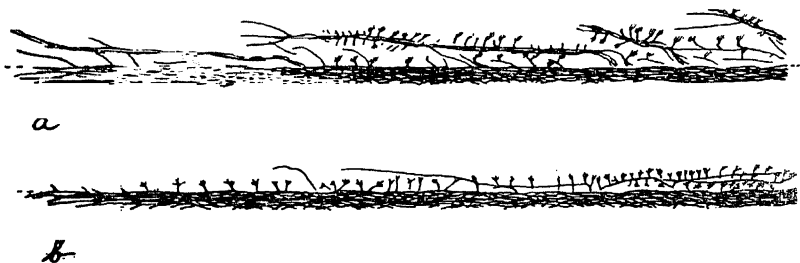


FIG. 92. *Scopulariopsis brevicaulis* (a) and var. *glabra* (b): Diagrammatic radial section (magnified 25 times) show the one culture (a) with prominent ropiness, the other (b) with only a few trailing hyphae.

The descriptions of species are arranged alphabetically since it is very doubtful if we know enough of the real relationships among them to arrange them more conveniently in any other manner.

481. *Scopulariopsis acremonium* Vuillemin. Bul. Soc. Mycol. France 27: 148. 1911.

Synonym: *Monilia acremonium* Delacroix. Bul. Soc. Mycol. France 13: p. 114, Pl. IX, fig. C. 1897.

Accepted by Oudemans for a culture in Arch. Neerl. Ser. II, T. 7, p. 285, Tab. XVIII, figs. 1 and 2. Oudemans' organism is regarded as differing from that of Delacroix and named *Scopulariopsis Oudemansii* by Vuillemin Bul. Soc. Mycol. France 27: 143. 1911.

Delacroix's Latin description may be translated: White, effused, slightly floccose; creeping hyphae hyaline, little septate, guttulate-granulate 4 to 5 $\mu$  in diameter; conidiophores erect, in groups (inter se propinquis), prominently septate, terminated by a chain of conidia; conidia ovate-pyriform, somewhat truncate at the base (and shown in the figure as having a basal pore), 12 to 15 by 8.5 to 10 $\mu$ , with a connective.

Species described from rotten paper, in Paris; cultivated by Delacroix without difficulty upon many substrata.

Delacroix's figure shows a section of a curving hypha bearing short 1- to 3-celled simple fertile branches arranged like a scorpioid cyme and each commonly tipped with one conidium; also a chain of conidia, and two conidia enlarged to show the thick wall and basal pore; conidia smooth, 12 to 15 by 8.5 to 10 with a very narrow basal ring and pore.

Unless some one can determine the organism back of such a description, none of these names can have any standing.

482. *Acaulium albo-nigrescens* Sopp. Monogr., p. 70-76, Taf. VI and VII, figs. 40-63. 1912.

Colonies chalk-white with coremium-like white hyphal bundles bearing conidial masses either on short conidiophores or sessile, and giving a mealy white appearance followed by the progressive development of black perithecia until the whole colony appears as a coal-black wrinkled mass of black bodies with areas of white showing among them; the underside is also black at this stage; conidiophores produced typically as branches from ropes, bundles, or coremia, forming *Stysanus*-like columns, branching of conidiophores in several superposed verticils with

elements progressively smaller in diameter; sterigmata long and tapering to a very narrow apex at the conidium-bearing tube, from which arise long diverging conidial chains; conidia white, recorded as 10 by  $10\mu$ , but figured as narrowly elliptical, possibly 10 by  $5\mu$ ; which become globose in germinating; perithecia arising as characteristic coils of hyphae involving the tips of several adjacent branches; olive-green then coal-black produced in great abundance ripening in several weeks and extruding their ascospore masses through fine pores as a pale coffee brown powder (ascospores) covering of the whole surface; some perithecia are superficial, others immersed in mycelial masses; ascospores about 6 by  $4\mu$ , fairly thick walled, oblique or concave on one side, brown, 8 to the ascus, extruded in slimy masses from the perithecia, and germinating slowly.

Species found first as a chalk-white growth upon milk in a cellar in Norway, and later in a compost-heap, and in old goat cheese.

Colonies grew between  $+1^{\circ}\text{C.}$  and  $35^{\circ}\text{C.}$  with optimum at  $25^{\circ}\text{C.}$ ; they grew well on gelatine and agar, poorly in rice, milk, beef-peptone broth, earth, and sawdust, richly on potato and bread, not at all on cellulose.

Biourge, *P. (Acaulium albonigrescens* (?) Sopp.) in Monogr. La Cellule 33: fasc. 1, p. 216-217, points out the inconsistencies in Sopp's discussion of his species and finds in his figures evidence that the organism had elliptical spores with long axis about twice the short axis, perhaps covered by the suggestion that the globose form indicated by Sopp's figures 10 by  $10\mu$  was reached in germination. Biourge suggests that the species belonged with the section containing *P. costantini*, *P. canescens*, *P. rufescens*, and *P. patulum* Bainier.

Biourge's culture labeled *P. albo-nigrescens* (our 4733.2) grew upon Czapek's solution agar as a submerged mass, rather hard, white then turning black suggestive of certain masses produced by *Cladosporium*. Conidial apparatus was not found in the mounts made but free spores that agree in general with the discussion were found.

483. *P. anomalum* Corda. Icones Fung. II, p. 18, Tab. XI, fig. 75. 1838.

Synonym: *Spicaria anomala* Harz, cited Sacc. Syll. 4: 167.

This fungus was described from rotting coniferous wood as delicate, white, with stipe erect, with an extensive tree-like branching system, the divergent tips bearing long curving chains of white elliptical conidia. Preparations of a form suggesting this description have been seen at the Cryptogamic Laboratory of Harvard University, but not successfully cultivated by us.

Corda's description reads: ? "*P. anomalum*: Tab. XI, fig. 75. tenuissimum, minutissimum, effusum, albidum; stipite erecto, supra ramoso, ramulis subregulariter positis, ramulisque brevibus flexuosis curvatis, adscendentibus laxis; floccis sporarum solitariis; sporis minutissimis ovatis. Long. spor 0.000160."

Enough question attaches to Biourge's identification of this species with *P. brevicaulis* to justify the recommendation that Corda's species be left forgotten.

484. *Acaulium anomalum* "ad interim" Sopp. Monogr., p. 65-67, Taf. VIII, fig. 75. 1912.

Synonym: *P. brevicaulis* Sacc.

Colonies at fruiting time white to ochre-brown, but growing poorly in laboratory media, mostly as submerged mycelium, irregular, often sterile and only tardily producing woolly masses of aerial hyphae and ropes of coremia with sparse areas of conidiophores, or sessile clusters of sterigmata and chains of conidia; reverse of colony and substratum weakly brown; conidia rough, angular, echinulate, brownish, 10 to 11 by  $8\mu$ ; perithecia not found.

Species obtained from Kral by Sopp as *P. brevicaulis* and later in various forms isolated in Norway; it failed to grow at  $+5^{\circ}\text{C}$ . and at  $40^{\circ}\text{C}$ .; it remained viable in the laboratory for ten years. Colonies grew best upon potato, poorly on rice, as sterile mycelia only on bread.

Sopp does not claim an identity for his organism with that of Corda since Corda is not mentioned. Its group identification is certain.

485. *Scopulariopsis arnoldi* (Mang. and Pat.) Vuill. Bul. Soc. Mycol. France 27: 137-152, generic change, p. 148, 1911.

Synonym: *Monilia arnoldi* Mangin and Patouillard.

486. *Scopulariopsis aureus* Sartory. Champignons paras., pp. 680-1, 1922. Described without name in Compt. Rend. Acad. Sci. 169: 703-4, 1919.

Grew equally well on potato, carrot and beef as well as peptone and sugar media, but did not grow on egg albumen and coagulated beef serum. Optimum temperature  $29^{\circ}$  to  $30^{\circ}\text{C}$ . with cessation of growth at  $39^{\circ}\text{C}$ . Mycelium in culture white then aureus, 0.5 to  $1.4\mu$  in diameter, branched; conidiophores erect, sometimes differentiated, tapering at the tip; conidia in chains, aureus, globose, ornate, 3 to  $4.5\mu$  (at  $37^{\circ}\text{C}$ .). In maltose or glucose gelatine there occur vesicular bodies, terminal or intercalary,  $130\mu$  in diameter.

A parasite of the human nails where it appears according to Sartory as irregular filaments, 2.5 to 9 or 10 $\mu$ , as terminal and intercalary bodies 20 to 35 $\mu$  in diameter and sometimes as conidia.

487. *P. Benzianum* Sacc. In Sylloge Fungorum 22: p. 1276. 1913.

Synonym: *P. insigne* Sacc. Ann. Mycologici 5: 178. 1907

Not *P. insigne* Bainier, and not *P. insigne* Winter.

Synonym: *Scopulariopsis* sp.

Colonies white then rosy, fairly compact, vegetative hyphae creeping septate, sparingly branched, 6 to 7 $\mu$  in diameter; conidiophores ascending, cylindrical, short 50 to 60 by 7 $\mu$ , sparingly septate, penicillus primary branch occasionally solitary usually both primary and secondary branches in threes, sterigmata with apex obtuse; conidia in very long chains, subglobose, rather large, 9 by 8 $\mu$ , smooth, minutely apiculate at the distal end, hyaline, in age pale rosy.

Species found upon moist and decaying leaves of *Citrus limo* at Pavia. Separated from *P. coccophilum* Sacc. by its smooth spores. Saccardo recognized the prior use of the name *P. insigne* and changed it in vol. 22 of the Sylloge repeating the Latin diagnosis of the previous paper without added information.

Judging by description this belonged in *Scopulariopsis* but no closer identification is possible.

488. *Scopulariopsis blochii* (Matruchot) Vuillemin.

Synonym: *Mastigocladium blochii* Matruchot Louis. In Compt. rend. acad. sci. (Paris) 152: 325-327, 1911. Un nouveau champignon pathogène pour l'homme.

On artificial media a filamentous, septate, colorless mycelium, 0.5 to 1.5 $\mu$  diameter developed; with conidiophores simple, tapering conical, 20 to 30 $\mu$ ; sterigmatic point bearing conidia in a centripetal manner; with conidia in chains, oval, with unequal ends, 3 to 4 by 1.5 to 2 $\mu$ ; in old cultures, white-creamy formations which may be undeveloped perithecia.

Found by Bruno Bloch in gummatous lymphangitis, having the clinical picture of sporotrichosis.

Consideration of Matruchot's original description together with Vuillemin's figure (Bul. Soc. Myc. France 27: 144-148, fig. on p. 145, 1911) indicates a general agreement in morphology with *Scopulariopsis*, but neither presents a clear conception of the conidium. Part of the sketches suggest a pointed apex and basal ring (or truncate base), while

others suggest monilia-like forms having both ends alike. The conidia as described are smaller than those found in the usual species of *Scopulariopsis*.

Etienne in 1920 in a medical journal from Nancy reports spore agglutination with *S. blochii* in summarizing studies on sero-reactions.

489. *P. brevicaulis* Sacc. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 219-220; Col. Pl. VI, Cart. 14, Pl. IX, fig. 54. 1923.

Synonyms: *P. anomalum* Corda, *Monilia Koningi* Oudemans, *S. brevicaulis* Bainier and *Acaulium anomalum* Sopp.

Colonies on wort gelatine, yellowish fulvous above and below, quickly liquefying the gelatine, with ammoniacal alliaceous or arsenical odor, no coremia in the strict sense; conidiophores 10 to 30 by 3 to 4 $\mu$ ; penicillus 20 to 40 $\mu$  long, all walls smooth, producing chains of conidia on single sterigmata or on any combination of sterigmata upon metulae or penicillate branching; branches crowded at base (insertae sedis), divaricate or none; metulae 8 to 17 by 2 to 3 $\mu$ , commonly in threes<sup>†</sup>; sterigmata 11 to 17 by 3 to 4 $\mu$ , in groups of 2 to 5; conidia lemonshaped, at first smooth, later echinate 5 to 9 by 4 to 7 $\mu$ .

Biourge no. 14 (our 4733.19) grew upon Czapek's solution agar as a gray colony becoming avellaneous in age, with abundant growth of superficial hyphae separate and in anastomosing ropes, forming a mass up to 1 to 2 mm. deep; with conidia 6 to 7 $\mu$  in long axis.

A pathogenic strain (no. 4858) of this group received from Dr. D. J. Davis of Chicago gave the following characters: Colonies upon Czapek's solution agar pale avellaneous spreading over the whole plate, with surface growth in coarse, anastomosing and more or less ascending ropes, becoming piled up to 5 to 8 mm. in depth in a zone halfway between the margin and the denser center; conidiophores irregularly produced as branches from the hyphal ropes and as very short branches scarcely rising above the substratum; sterigmata up to 30 $\mu$  long tapering slowly to an apex about 3 $\mu$  in diameter; conidia 6 to 8 by 5 to 6 $\mu$ , rough, pale, avellaneous, with basal ring about 3 $\mu$  in diameter, in long chains.

490. *S. brevicaulis*, var. *alba* Thom.

Synonym: *P. brevicaulis* Saccardo, var. *album* Thom. U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, 47, fig. 13. 1910. See fig. 93.

Colonies upon sugar gelatine white to cream-colored alike above and below upon all media, strict to sparsely floccose, with trailing hyphae and ropes of hyphae, indeterminately spreading; conidiophores either

arising from substratum directly or mostly as perpendicular branches of aerial hyphae and ropes of hyphae 15 to 40  $\mu$  in length, penicillus varying from a single chain to more or less complex penicillate branching, mostly producing few chains of indefinite length and arrangement from narrow tapering sterigmata; conidia pyriform to subglobose, with basal collar, 9 to 10  $\mu$ , roughly tuberculate, white or slightly yellowish tinged, thick walled except at the base, the center of which remains as a germ pore. Colonies rapidly liquefy sugar gelatin with strong ammoniacal odor, and give an intensely alkaline reaction in litmus media. Gives exactly the same reactions as *P. brevicaulis* Sacc. Differs from the latter slightly, except in the color of the spores.

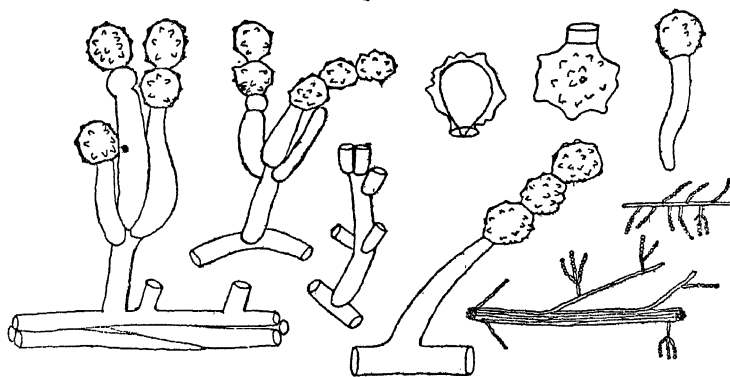


FIG. 93. *Scopulariopsis brevicaulis* var. *album* Thom: Figures from 1910 paper, show only minor differences from the species.

Common upon imported Camembert cheese. Found often upon domestic Camembert and grows very readily in cheese cellars, where it becomes a nuisance.

490a. *P. brevicaulis* var. *album* Thom. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 226-227; Col. Pl. XII, Cart. 372; Pl. XX, fig. 118. 1923.

Biourge found this form to produce a pseudoparenchyma of large cells, to produce ascospores smooth without furrow, but did not find the perithecia. He decided that Thom's nomenclature was not valid but offered no change.

The figures and information given do not, however, indicate that Biourge ever identified Thom's no. 4. Hence his findings cannot be



assigned to that organism; the culture was listed as no. 4733.44 in our collection.

490a. *Scopulariopsis brevicaulis* var. *glabra* Thom. In U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 48-49, fig. 16. 1910. Described as *Penicillum*.

Colonies white or only slightly yellowish-tinged in all gelatin media, grow not at all or with difficulty on agar of most formulae. Aerial portion consisting of short, closely crowded conidiophores making a powdery surface overgrown by loosely trailing hyphae and ropes of hyphae, spreading broadly over the substratum. Conidiophores, short, mostly 10 to 30 $\mu$ , arising directly from submerged hyphae or numerous and irregularly borne as perpendicular branches of the superficial hyphae and ropes of hyphae. Penicilli from simple chains of spores to fairly complex penicillate groups of branchlets resembling *S. brevicaulis*, but mostly less complex. Conidia obovate, pyriform 7 to 8 by 8 to 10 $\mu$  or almost globose, 7 to 9 $\mu$ , smooth, white, rather thick-walled and retaining their power to germinate for many months. In old potato and other cultures black sclerotia are formed in the substratum but do not produce asci. Liquefies gelatin rapidly (within one week), gives a strong alkaline reaction and ammoniacal odor.

Habitat: found repeatedly on imported Camembert cheese and secondarily upon domestic soft cheese, where it grows into prominent cottony patches indistinguishable to the eye from the white variety of *S. brevicaulis*. This fungus is separated from the species by its smooth white spores and the production of the black sclerotia in the substratum.

This form is certainly closely related to *S. brevicaulis* by its physiological reactions and its general morphology. It was found in one case among the exsiccati in the Harvard herbarium under the name of *Monilia candida*. There is, however, no possibility of confusing this form with that species as understood and described by more recent students, such as Hansen and Jorgenson.

Cultural data: Exactly as in *P. brevicaulis*, except for the following: Conidia smooth, somewhat smaller; color more nearly white; sclerotia black, found in very old potato-plug cultures or in agar cultures which have grown several weeks or months. Cohn's solution failed to produce a characteristic colony.

*Oospora glabra* Hanzawa (Hanzawa Jun. Unters. ü. Pilze auf dem Getrockneten Boniten oder Katsuobushi. In Jour. Coll. Agr. Tohoku Imp. Univ. Sapporo 4 (1911) Pt. 5, p. 238, Pl. XXIII, figs. 1-6) is

probably this same species. It is quite possible that *P. ovoideum* Preuss was also this species.

One reference to *P. brevicaulis* var. *glaucum* has been seen but it is probably a misprint for this form since no green species is known in this group. •

491. *Scopulariopsis brevicaulis*, var. *hominis* Brumpt and Langeron.

Brumpt, E., Précis de Parasitologie, 2d ed., p. 902-5, 1913.

Synonyms: *Penicillium brevicaulis*, var. *hominis* Brumpt and Langeron, Brumpt, Précis de paras., 1st ed., p. 838-840, figs. 652-3, 1910.

Colonies on glycerinated potato, sweet potato, carrot, glucose, maltose, and peptone medium of Sabouraud, velvety, forming vesicles either intercalary or terminal, up to  $115\mu$  in diameter; hyphae on surface of medium in tufts, aggregated and in coremia, (in ropes?); conidia cocoa brown, (café au lait, according to Émile-Weil and Gaudin) ornate, spherical, sometimes lemon shaped.

This variety was found in two cases of onychomycosis of the toes by Brumpt and seven cases by Émile-Weil and Gaudin (p. 456-8, fig. 1). Brumpt in collaboration with Langeron found in the original material from the infected nails, mycelial filaments, 2 to  $10\mu$  in diameter, septate; and chlamydospores, terminal and intercalary, 10 to  $30\mu$ . Émile-Weil and Gaudin in addition report conidia. The parasitized parts of the nail are said to be much darkened, often a dull yellow brown.

These strains as described are not to be distinguished from others secured from diseased areas and designated by specific names.

492. *P. brevicaulis* Sacc. forma *intermedium* D. Sacc. (Mycotheca italica 1726. Centurie XVII, Padua, prior to May 1913). Printed label is signed G. Gagnetto.

Cultured on agar conidia catenulate, subglobose, smooth, asperulate at one end, 8 to 9 by 5.5 to  $8\mu$ , avellaneous.

Habitat: Body of a woman who died from arsenical poisoning. An intermediate form of *P. brevicaulis* var. *glabrum* Thom.

Examination of the herbarium material shows the *Scopulariopsis* type of spore, catenulate, delicately rough, very apiculate, and distinctly truncate, thick walled, pale brownish, 6 to 9 by 5 to  $7.5\mu$ . Many immature spores were noted. The specimen was mounted for examination in KOH.

This form name may be disregarded as the conidia are not smooth when mature as stated on the label accompanying the specimen.

493. *Penicillium brevipes* Sacc. (incorrectly labeled for *P. brevicaulis*?). Patouillard collection, Farlow Herbarium "Sur Criquet peleron. Olgerde. 1899. Trabut leg."

The specimen is a portion of a cricket collected in 1899 on which is a pinkish fawn powder. The conidia composing the powder were of the *Scopulariopsis* type. See no. 637 for *P. brevipes* Corda.

494. *Scopulariopsis candelabrum* Loubière. (See Loubière, A. Thèse présenté a la faculté des sciences de Paris, Serie A, no. 982, no. d'ordre 1812. 1924. Recherches sur quelques Mucedinees caseicoles, pp. 93, Pls. 9. Diagnosis p. 63-64, Pl. VII, figs. 13, 14.

Cultures upon neutral and salted media white, floccose, becoming later compact, tuber-like in appearance from the mass of powdery white spores; sterile hyphae creeping, branched, tangled, septate, 2.5 to 3 $\mu$  diameter; conidiophores erect, branched, septate, up to 40 to 60 $\mu$  long and from 4 $\mu$  at base to 2 $\mu$  diameter at apex, with 3 or 4 branches in pairs (see figure) below septa incurved and narrowed toward the tip, the whole looking like a candelabrum; conidia in chains, with delicate connective, often 40 to the chain, about 8 to 10 $\mu$  in diameter figured as smooth.

495. *Scopulariopsis candida* (Gueguen) Vuill. Bull. Soc. Myc. France 27: 143. 1911.

Synonym: *Monilia candida* Gueguen 1899, not of Bonorden.

- 495a. *S. candida* (Pers.) Loubière. Thèses présentées à la Faculté des Sciences de Paris, Sér. A, no. 982, no. d'ordre 1812, p. 64, 65, 73, Pl. VIII, IX. 1924.

Suggested synonyms: *Monilia candida* Persoon 1822; *Aspergillus candidus* Link, *Sterigmatocystis candida* Sacc. Ascosporic form: *Nephrospora Mangini* Loubière.

See *S. candida* (Gueguen) Vuillemin no. 495. Identification with Persoon's species may be regarded as doubtful as also identification with any *Aspergillus*.

Loubière figured and described *S. candida* as the conidial stage of his genus *Nephrospora Mangini* Loubière (citation as above). In the *Scopulariopsis* form the hyphae were uncolored; sterile trailing hyphae produced conidia-bearing branches of varying length with penicilli vary-

ing from single sterigmata with one chain of hyaline conidia about 7.5 by  $6\mu$ , figured as smooth, to various clusters of sterigmata and conidial chains. He described perithecium formation in *Nephrospora* as resulting in pyriform subcarbonaceous bodies 90 to 175 by 85 to  $150\mu$ , with ostioles; asci about  $10\mu$  in diameter; ascospores 5 by  $3.5\mu$ , yellow or golden yellow, and kidney-shaped.

Manifestly Loubière's description raises the question whether other species of *Scopulariopsis* belong also in *Nephrospora*. At present no answer can be given. It also raises the question whether Sopp's *Acaulium albo-nigrescens* (see no. 482) may not have been this species since the descriptions and measurements have much in common.

496. *Scopulariopsis casei* Loubière (see Loubière, A. I. Thèse Recherches sur quelques mucédinées caseicoles. Thèses présentées à la Faculté des Sciences de Paris Serie A, no. 982, no. d'ordre 1812, 1924), diagnosis, p. 62, pl. VII, fig. 12.

Grown upon the usual media, colonies are comparatively thick (epaisse) compact and white, later becoming a clear tint of yellow.

Mycelial hyphae uncolored, septate, branching, anastomosing, and sometimes coiled tendril-like, usually 2 to  $3\mu$  in diameter; conidiophores erect, unicellular, always simple (unbranched ?) and scarcely larger than the vegetative hyphae, 5 to  $15\mu$  long, each bearing a chain of conidia separated by delicate connectives (disjuncter), and commonly up to 20 conidia to the chain; conidia somewhat ovoid, somewhat rounded at the apex and flattened at the base, about 6 to  $7\mu$  in diameter, echinulate and a very pale yellow.

This species approaches *S. acremonium* (Delacr.) Vuill. and *S. Arnoldi* (Mang. et Pat.) Vuill., especially the latter, and probably stands between these two species.

497. *Scopulariopsis castellanii* Ota and Komaya. Über eine neue Art der Gattung *Scopulariopsis* (Bainier): *S. Castellanii*. Dermatolog. Wochenschr. 78: 163-165, Abb. 1. 1924.

On dextrose and beer wort agar greyish white, later white, with ropes, powdery. Hanging drop cultures mycelium  $5\mu$  in diameter septate; conidiophores upright, 3 to  $4\mu$  in diameter, 2 to  $40\mu$  in length, unbranched or dichotomously branched, septate; sterigmata (?—C. T.) thick rounded (probably youngest conidium); conidia truncate, oval, tuberculate when mature, with a thick membranous outer wall, 7 to  $9\mu$  in diameter, catenulate, often separated by an isthmuslike connective. No perithecia observed.

Original secured from Castellani in Colombo by Fulleborn and Meyer in 1906. Maintained in culture by Nauck for eighteen years.

The describers found this strain non-pathogenic for the guinea pig, but pathogenic for two white mice. A suspension of the organism introduced intraperitoneally caused death in fourteen and nineteen days. On post-mortem typical conidia were found by microscopic examination in an abscess on the inner surface of the peritoneum, in the mesenteric lymph glands, the kidneys, liver and spleen, but no mycelia and conidiophores.

498. *Scopulariopsis cinerea* Emile-Weil and Gaudin. Arch. Med. Exp. et Anat. Path., Paris 28: 458-460, Text-fig. 2, and Pl. 12, fig. 2.

Colonies on carrot, velvety, white, ashy gray, finally brown green, both mycelium and conidia becoming brown in age; hyphae united into coremia (trailing ropes?); on maltose gelatine of Sabouraud becoming entirely green with age; conidia with truncate base and pointed apex, 4 to 5 by 2.5 to 3 $\mu$ ; chlamydospores abundant both terminal and intercalary, up to 100 $\mu$ .

Perithecia developed in about three weeks at 25°C. beginning as the coiled tip of a hypha (spiral), then surrounded by pseudo-parenchyma finally becoming black, globose, about 250 $\mu$  in diameter and surrounded by radiating mycelium; asci 8-spored, about 10 to 12 by 8 $\mu$ , commonly breaking down to leave the ascospores free in the perithecium; ascospores brown, plano-convex, 6 to 7 by 3 to 3.5 $\mu$ .

Species found in several cases of infected great toe.

In the infected nail appearing as knotty filaments, 2 to 4 $\mu$  in diameter as terminal and intercalary chlamydospores, 15 to 20 $\mu$ ; and rarely as brown mycelium terminating in brown conidia.

The description of the conidia relate this strain to the genus assigned. The colors described appear to be based upon reactions in the substratum not upon the masses of conidia. No color due to conidial masses is specified in the description.

499. *P. coccophilum* Sacc. Ann. Mycol. 5 (1907), p. 178.

Saccardo's Latin description may be cited in full: "Effusum, parasiticum, carneolo-isabellinum, densiuscule mucedineum, hyphis sterilibus repentibus, pareis; fertilibus seu conidiophoris adscendentibus, brevibus, tatis (cum ramis) 90-120 $\mu$  altis, 5.5-6 $\mu$  cr., parce septatis; ramis arrecto-penicillatis, imis oppositis v. solitariis, superioribus bis 3-4 verticillatis, ultimis sensim sursum tenuatis; conidiis globosis v. subglobosis, exquis-

ite verruculosis, majusculis,  $8-9 \times 8$  carneolo-isabellinus, catenulatis. Hab. cum prœcedente *Stilbo coccophilo* et partier ut videtur parasiticum."

This species was described from preserved specimens in terms which suggest a member of the *P. brevicaulis* or *Scopulariopsis* group. It was not cultivated and unless restudied under conditions approximating those of the original material, cannot be surely identified. Conidia 8 to 9 by  $8\mu$ , isabelline in color and verruculose, place it fairly closely with related forms already described.

500. *P. coffeicolor* B. and Br. Ann. N. H. n. 1614.

Possibly a *Scopulariopsis*.

The only significant data given are colonies umber, conidia globose 12 to  $13\mu$  in diameter.

Cultures: On Pasteur's solution. Britain, not again reported.

501. *Scopulariopsis communis* Bainier. Bul. Soc. Mycol. France 23: 125-127; Pl. XVI, fig. 3-6. 1907.

Synonym: *P. scopulariopsis* Sacc. Sylloge 22: 1275, no. 8033.

Colonies upon licorice sticks producing prominent ropes and networks of hyphae almost perpendicular to the substratum, and bearing abundant conidial fructifications as branches; penicillus figured very short (1-celled) stalk, bearing a verticil to sterigmata directly or a mixed verticil of metulae and sterigmata, or more complexly branching; conidia more or less oval, with truncated base and pointed apex, about 5.6 to 11.2 by  $3.6\mu$ , almost colorless at first becoming cream when ripe.

Culture no. 2729 (lost from our collection) from Miss Dale reproduced Bainier's description and was so reported by her (Ann. Mycol. 12:46, Pl. IV, fig. 71, 72, 1914) with confirmation by Bainier. Saccardo merely transferred the species to *Penicillium* changing the name and compiling a Latin diagnosis.

501a. *Scopulariopsis communis* Bainier. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 217-218; Col. Pl. VI, Cart. 10, and XII, 363; Pl. IX, fig. 53 and XX, fig. 117. 1923.

Synonym: *Isaria casei* Mazé under which Biourge places his cartons.

Colony on wort gelatine hyaline, commonly submerged, rapidly liquefying, with ammoniacal odor, also arsenical or onion-like odors, producing (coremia) ropes of hyphae commonly sterile only occasionally conidia-bearing; conidiophores very short, 4 to  $5\mu$  in diameter, sometimes

undivided and producing conidia directly with all walls smooth; metulae rare 10 to 14 $\mu$ ; sterigmata mostly deformed (irregular), 20 to 25 by 1 to 5 $\mu$ , tapering into undulate beaks; conidia 7 to 10 by 6 to 10 $\mu$ , globose or truncate at base.

Biourge's no. 10 and no. 363 (not received by us) as described differ considerably in measurements from Bainier's organism but since his cultures have not been studied no attempt to place them accurately will be offered.

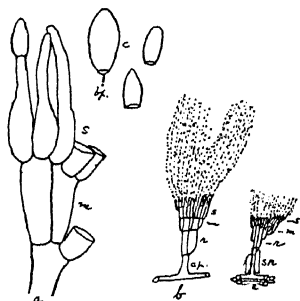


FIG. 94. *S. Costantini* (Bainier) Dale: Our figures showed conidia, *c*, with a basal ring and pore, *bp*; supporting branches, *sr*, turning backward from about the middle of the short conidiophore, *cp*. These branches part directed upward and part downward sometimes formed an apparent coremium at the base of a very large penicillus.

502. *Scopulariopsis costantini* (Bainier) Dale Ann. Mycol. 12: 57, 1914.

Synonym: *P. costantini* Bainier, Bul. Soc. Mycol. France 22:

205-207, Pl. XI, fig. 1-6, 1906 (compare our fig. 94). *Synpeni-*

*cillium album* Costantin, Bul. Soc. Mycol. France 4: pp. 62-68.

Pl. XIV. figs. 10-17, 1888.

Colonies upon licorice sticks white, producing abundant aerial mycelium up to 10 mm. in depth in the form of trailing and ascending hyphae and anastomosing ropes of hyphae, appearing mostly as separate hyphae at the margin; conidiophores arising as branches and even bundles of branches from the aerial mycelium, coarse, short, several-septate, figured as showing both the apical penicillus and reversed appressed branches toward the base and running back to mycelium; penicillus figured as a complex system of appressed branches from 1 to 3 upper nodes each terminating in a verticil of metulae sterigmata and chains of conidia in compact masses, often with several penicilli combined and interwoven; conidia elliptical.

Species recorded by Bainier as common on straw, wet paper and dung in France. A culture under this name was received from Biourge (no. 4733.44) but proved to be *P. brevicaulis* var. *glabrum* Thom; cultures identified by us with Bainier's description are 2744 (not now viable) from Miss Dale obtained from soil, and 4701.76 from Mrs. Kidd in England.

From cultures of these strains on Czapek's solution agar the following additional data are drawn:

Colonies at first pale yellowish toward orange, thin at edges and indistinctly zonate then close velvety, in older areas overgrown with interlacing ropes of hyphae bearing conidiophores; conidiophores about  $60\mu$  from base to penicillus, stalk, branches and metulae coarsely tuberculate; branch or branches up to  $20\mu$  by  $4\mu$ ; metulae about 10 by  $4\mu$ ; sterigmata up to 10 to 12 by 2 to  $2.5\mu$  closely packed in the verticil, acuminate pointed; conidia colorless elliptical, thick walled, smooth 3.5 to 6 by 2 to  $3.5\mu$ , with careful examination, seen to have apex more or less pointed and the ring or collar at the base allying it with *P. brevicaulis* or Scopulariopsis, hence the placing of the species.

Bainier properly discards Costantini's genus *Synpenicillium* as untenable. If the occurrence of ropes of hyphae producing conidiophores as short branches were accepted as the basis for a genus, it would arbitrarily take members of the penicillium group from various sections to which they are clearly related and place them in a heterogeneous group. The close association of Bainier with the Paris group of Mycologists probably justifies accepting the identities claimed.

When Miss Dale sent us her culture no. E16 it was recognized as *P. constantini* Bainier and so reported to her. Later she informed us that the identification had been verified by Bainier. From our observation of a germinal pore at the base of the conidium we placed the species with the Scopulariopsis series as Miss Dale published it although Bainier had reported it as a redescription of *Synpenicillium album* of Costantini which was the type of Costantini's genus. The presence of this germinal pore is not reported by Bainier in any of these species and was not observed by us in those of the series we have had in culture. We have left *P. constantini* with Scopulariopsis although morphologically it seems to belong with the polyverticillate series. Biourge may be correct in regarding them all as more closely related to Scopulariopsis than to the true Penicillia.

Gelatine in water was liquefied, without discoloration, with the odor of *P. brevicaulis* Saccardo.



Bean agar produced much richer growth than Czapek's solution agar, white or tinged very slightly yellowish, especially below, with much heavier growth and more tufted appearance. The morphology of this species together with its reactions in gelatin show close relationship to *P. brevicaulis* which formed the type of Bainier's genus *Scopulariopsis*. On the other hand, the penicillate conidial fructifications produced lead Bainier himself to overlook this relationship and call it *Penicillium*. Cultivated by Miss Dale, Cambridge, England, from soil.

502a. *P. constanti* Bainier.

A misspelling in Trans. British Myc. Soc. 5 (1914): 163. 1915, Smith and Ramsbottom.

503. *Oospora cretacea* (?) Harz. In Biourge Monogr. La Cellule 33: fasc. 1, p. 229; Col. Pl. XII, Cart. 300; Pl. XXI, fig. 124. 1923.

Colonies in wort gelatine, thin and easily broken, white or later more or less salmon; reverse yellow to fulvous; odor arsenical or alliaceous; gelatin liquefied; conidiophores about  $3\mu$  in diameter decumbent; sterigmata 10 by  $3\mu$  when present but rarely produced; conidia angular-globose about  $5\mu$  in diameter, in a "continuous or discontinuous" series, at the branching apex of a stalk or upon true sterigmata.

Biourge's no. 300 (our no. 4733.45) is noted as certainly belonging to the series with Harz's species whether it is identical or not. Colonies in Czapek's solution agar were white producing an interlacing overgrowth of hyphae about 100 to  $200\mu$  deep, with reverse yellow to deep orange; on wort bristly masses rising above the surface.

This appears to be another doubtful member of the group.

504. *P. divaricatum* (?) Thom. In Biourge Monogr. La Cellule 33: fasc. 1, pp. 224-225; Col. Pl. VI, Cart. 83; Pl. X, fig. 59. 1923.

Misidentification of some variety of *P. brevicaulis*.

Biourge's culture no. 83 (our no. 4733.52) confirms his figure in putting his organism with the *P. brevicaulis* series. He accepts the name on a culture received from Kral as representing Thom's species while he had Thom's organism or one of its varieties in his hands as *P. aureocinnamomeum*.

Our own material purchased from Pribram later as representing the old Kral collection contained our no. 4777.10 labeled *P. divaricatum* but actually *P. brevicaulis*.

507. *Scopulariopsis fimicola* (Cost. and Matr.) Vuillemin. Bul. Soc. Mycol. France 27: 137-152; generic change, p. 143, 1911.  
Synonym: *Monilia fimicola* Costantin and Matruchot, 1894.  
Apparently the change is purely bibliographic.

508. *Acaulium flavum* Sopp. Monogr., p. 53-56, Taf. IX and XI, figs. 76 to 79. 1912.

Colonies white with a yellowish red tint (? avellaneous?) and coarse septate, branching hyphae, which are rich in fat, at first submerged but quickly developing aerial growth; in gelatin conidial development comes slowly; after several weeks the colonies have a dry powdery or mealy white appearance with a yellowish red tinge; conidiophores often wanting, very short or forming long stalks, with *Penicillium*-like branching only in young cultures, while the stalk is commonly entirely suppressed in old cultures, with 1 to 4 series of branches; sterigmata sometimes sessile upon mycelium, again branched or septate, long curved, pointed; conidia somewhat pyriform, colorless or leather yellow, rough or echinulate, or angular, or at times smooth, about 7 to 8 $\mu$  in diameter; perithecia small black-green, imbedded in yellow sclerotium-like masses of mycelium, only produced upon potato in Sopp's cultures, produced first as yellowish sclerotia densely covered with conidial masses, within which the perithecia at first appear as yellowish red then greenish black masses, which develop very slowly and were not fully described.

The species was found upon dead insect larvae in western, eastern and southern Norway, also on cheese-rinds, especially on goat-cheese and on sour whey cheese. In culture, the optimum temperature was about 25°C.; growth was poor at 35°, stopped at 40°C., but persisted down to 3°C.

No one has since identified it hence the data as to ascospore formation remains incomplete.

509. *Acaulium fulvum* Sopp. Monogr., p. 67-70, Taf. IX, fig. 81-84; Taf. XII, fig. 80. 1912. Possibly described as an *Isaria*.

Colonies *Isaria*-like, white, yellowish or reddish, with *Stysanus*-like coremia, growing in culture as a close but rather thin felt over the substratum, white, or gray above and in reverse, becoming yellowish with a reddish tinge with the ripening of conidia; hyphae fairly coarse, *Mucor*-like, septate, branched, thin-walled, with few vacuoles; conidiophores commonly branches from prostrate ropes of hyphae, fairly typically

penicillate in their branching, varying from 1 to several cells in length or wanting; sterigmata as sessile upon mycelial bundles, or in penicillate verticils; conidia smooth, yellowish, 10 to 14 by  $5\mu$ , produced as in *P. digitatum* Sacc., hence initially cylindrical, then oval, chlamydospores or round oidia produced directly from the mycelium or in short branches, as irregularly globose cells much larger than the conidia and filled with fat; perithecia not found.

Species found upon dead insects and their excrement, where thin white and reddish coremia are visible with the naked eye.

The description of conidium formation would exclude this from the group if correct and place it with *P. divaricatum* and Sopp's *Corollium*.

511. *S. insectivora* (Sopp). Biourge Monogr. List.

Synonym: *Acaulium insectivorum* Sopp. Monogr., pp. 60-64, Taf. IV and VIII, figs. 66-69. 1912.

Colonies clear yellow-brown to reddish ocher-brown, varying somewhat with the substratum, with surface growth dry, mealy, knotty, bristly, or showing coremia; mycelium coarse, at first submerged like *Oidium lactis*, but soon producing conidia over the whole surface giving a mealy or powdery and wrinkled rather than a fibrous appearance; reverse whitish showing clumps like sclerotia in age; conidiophores irregular, sometimes reduced until the conidial apparatus appears to be sessile, sometimes long stalked, like typical *Penicillia*; branching either present or wanting, often forming a penicillate system 50 to  $100\mu$  long; sterigmata commonly in dense verticils which are often sessile on the mycelium in old cultures; conidia 10 to 11 by  $9\mu$ , (in discussion p. 61 3-4 by  $5\mu$  ?) somewhat yellowish, more or less oval, thick walled, at first smooth, but in old cultures angular, rough, almost echinulate; perithecia not found; sclerotium-like pseudo-parenchymatous masses are found in old cultures.

Species abundant in Norway, upon dead flies and larvae, in earth upon decaying vegetables, fibers, found upon caraway seed which few fungi infect, a common contaminant of laboratory cultures; colonies grow between  $+1^{\circ}$  and  $+40^{\circ}\text{C}$ . with optimum  $30^{\circ}$  to  $35^{\circ}\text{C}$ ., and flourish in a wide range of media and with a considerable range of physico-chemical characters as reported by Sopp.

Although we bought a culture from Pribram under this name (no. 4777.25) and found it a *Scopulariopsis* we doubt its authenticity since it does not comply in any close degree with Sopp's description.

512. *Scopulariopsis ivorensis* Boucher. Bull. Soc. Path. Exot. 11, 309-315. 1918.

Colonies described as clear to deep chestnut to deep brown spreading radiately by a white border of aerial hyphae; hyphae  $1\mu$  in diameter; conidiophores short, irregular with long sterigmata; conidia about  $6\mu$  in long axis, brownish, rough, with truncate base.

Isolated from lesions over bony areas upon an African. Regarded as a variety by Boucher but described as a species.

513. *Scopulariopsis koningii* (Oudemans) Vuillemin. Bul. Soc. Mycol. France 27: 143. 1911.

Synonym: *Monilia koningi* Oudemans. Arch. Neerland. Sc. Exact. et Nat. p. 23. Tab. XXI. 1902.

The significant portion of Oudemans' Latin diagnosis may be translated: Colonies on soil extract gelatine orbicular, subzonate, rosy avellaneous; hyphae all hyaline 4 to  $5\mu$  in diameter, septate; the creeping hyphae dichotomously branched; ascending hyphae racemously branched with branches (sterigmata ?) basidium-like, lageniform, 30 to  $40\mu$  long, bearing single chains of conidia; conidia up to 20 in the chain, subglobose, apiculate at the apex, smooth, 6 to  $8\mu$  in diameter, delicately rosy avellaneous.

Oudemans described the species from Koning's culture obtained from humus or leaf mold in a forest near Bussum, Holland. His figure clearly identifies his organism as a smooth spored member of the *P. brevicaulis* or *Scopulariopsis* series. The species has not been identified by us. Culture no. 4500.341 received under the name has conidia 7 to  $8\mu$  in long axis, but has rough points.

Castellani and Chalmers (Manual of Tropical Medicine, Aldo Castellani and A. J. Chalmers, 2d ed., 1913, London, p. 841-2) suggest as synonymy: *Monilia koningii* Oudemans, *Scopulariopsis refulus* Bainier, *S. koningii* Vuillemin, 1912.

514. *P. "Linguae (genre Scopulariopsis)."* Panayotatou in Centralb. f. Bakt. etc., 1 Abt. Orig. 101: Heft. 4/5, pp. 231-235; text figures 1-6. 1927.

The one significant item in the discussion of this mold is a reference that Langeron had identified it as belonging in *Scopulariopsis*.

The conidia are given as 3 to  $6\mu$  in diameter, the colonies as greenish to brown wrinkled.

Habitat: Isolated from lesions on a child's tongue in Alexandria, Egypt. (See Chapter IX.)

515. *Acaulium nigrum* Sopp. Monogr., p. 47-53, Taf. X, figs. 86-89; Taf. XI, fig. 85, 92, 93; Taf. XII, fig. 90, 91. 1912.

Colonies brown toward black, color potato and gelatine blue-black, in gelatine cultures remaining long submerged in the liquefied brown mass, ultimately producing conidial areas about the margin and over most of the surface which is irregular or rough and wrinkled; in very old colonies successive new growths overlay each other to form deep masses; hyphae delicate upon the usual media almost *Streptothrix*-like; conidiophores often wanting, occasionally present and *Penicillium*-like, especially in young cultures upon potato, branching at the summit 1, 2 or 3 times to produce a tangled often winding mass of sterigmata, bearing long chains of conidia; sterigmata frequently sessile on mycelial hyphae, especially in old cultures; conidia large, irregularly echinate, angular, almost polygonal, thick walled, at first violet brown then various shades of red, chocolate and coffee brown to black-brown, 7 to 8 $\mu$  diameter; perithecia produced most readily on potato and in the spring, at first olive green, then black, sunk in the mycelium, with definite ostiole; ascospores eight to the ascus, smooth, oval, almost sharp pointed, brown, 7 by 5 $\mu$ ; sclerotia variously produced, forming tough cartilaginous masses 3 to 5 mm. thick and up to 10 by 20 mm. in diameter but produced only when conidium formation is suppressed, another type forming coarse white masses and a third type forming black masses 250 $\mu$  are listed.

The species was parasitic upon insect larvae especially *Gastropacha pini* and recoverable from earth samples in the infected area; it was reported from various places in Norway; upon the larvae, colonies form a thin close woven mycelial layer, which is densely covered with sessile sterigmata.

In describing the conidia (p. 49) Sopp fails to note the characteristic basal ring and pore, but refers to some of the conidia as pointed at both ends.

*Cultural characters* (as given by Sopp): Colonies grew poorly on gelatine media with mycelium mostly submerged and few conidia, although the gelatine was liquefied and brown; upon agar growth was more favorable and sclerotium production evident; growth in milk was slow, in urine rapid, in beef-peptone broth slow; in broth with 1 per cent tartaric acid it stopped. Wort was a favorable medium and potato was very favorable to the production of rich growth and perithecia; similarly bread produced good colonies. Spores retained their vitality for 5 years in storage.

No one else has reported this species.

516. *Scopulariopsis penicillioides* (Delacr.). Smith and Ramsbottom, in Trans. British Mycol. Soc. 5 (1914): 164. 1915.

*Monilia penicillioides* Delacroix. Bul. Soc. Mycol. France 13: 114-115. Pl. IX, fig. B<sub>1,2,3</sub>. 1897.

*P. penicillioides* (Delacr.) Vuillemin. Bul. Soc. Mycol. France 27: 75-6. 1911.

Delacroix's diagnosis translated freely: Colonies at first white, effused, sublanose, from cinereus to yellowish (pale café au lait color), powdery; conidiophores erect, sometimes simple mostly umbellately branched at the apex, with ultimate branchlets rather obtuse at the apex and bearing chains of conidia; conidia hyaline, somewhat yellowish in mass, broadly lemonshaped, smooth at first, echinulate when ripe with distal end pointed, with connective, 6 to 7 by 4.5 $\mu$ .

In Delacroix's figures single sterigmata and short penicillate branches are irregularly arranged upon trailing hyphae, with the sterigmata either uniform in diameter from end to end or with long tapering points producing conidia as in *Scopulariopsis* but without showing the structure of the conidium characteristically. Species found upon crickets dead in the field. Parasitism was suspected but not proved. Cultures grew readily upon various culture media.

Smith and Ramsbottom place under this name the culture described by Miss Dale as *M. Koningi* (Ann. Mycol. 19: 460, 1912) and later as *S. rufulus* (Ann. Mycol. 12: 45, 1914). It is doubtful if any one knows or will determine exactly which form was studied by Delacroix.

517. *Scopulariopsis repens* Bainier, Bul. Soc. Mycol. France 23: 125-127; Pl. XVI, fig. 1-2, 1907.

Synonym: *P. bainieri* Sacc. Sylloge 22: 1275, no. 8034. (See Fig. 95).

Colonies not described except in the terms applicable to the group; conidiophores reduced to 1 or 2-celled branches perpendicular to trailing or ascending hyphae and ropes of hyphae; conidial apparatus consisting of sterigmata partly isolated, partly in groups or verticils of 2 to 6, and partly with verticils grouped into short stalked penicillus-like masses variously distributed along the hyphae; branches or metulae partly cylindrical, partly short and variously swollen; sterigmata 14 to 28.5 $\mu$ , diverging at the apex and producing long diverging chains of conidia; conidia globose 5.6 to 8.4 $\mu$  in diameter, delicately echinulate, pale near "café au lait."

Saccardo made a routine transfer of this species as *P. bainieri* to *Penicillium* and summarized Bainier's description into a Latin diagnosis.

517a. *P. (Scopulariopsis) repens* (Bainier). Biourge Monogr. La Cellule 33: fasc. 1, pp. 225-226; Col. Pl. XII, Cart. 362; Pl. XX, fig. 119. 1923.

Colonies on wort gelatine, frequently forming a pseudoparenchyma, tardily producing aerial growth, usually almost colorless or sordid rosy,

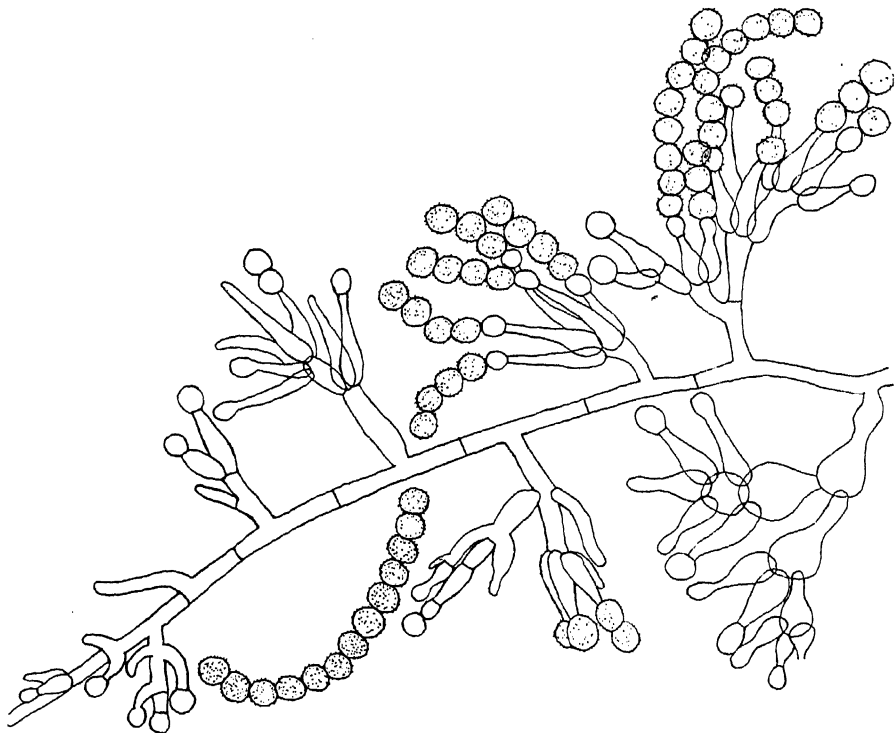


FIG. 95. *S. repens* Bainier: Bainier's beautiful figure is used here to show the type of structure; apparently Bainier never fully understood the characteristic structure of the conidium in the group.

reverse yellow; gelatine quickly liquefied; odor alliaceous or arsenical strong; conidiophore 6 to 7 $\mu$  in diameter; penicillus, if the term may be used, about 35 $\mu$  long; sterigmata 15 to 20 by 5 to 7 $\mu$ , either very long with bristle-like tube or none! conidia globose 9 to 10 $\mu$ .

Biourge's no. 362 (our no. 4733.101).

518. *P. (Oospora) rosatum* Biourge, Monogr. La Cellule 33: fasc. 1, p. 228–229; Col. Pl. XII, Cart. 393; Pl. XXI, fig. 123. 1923.

Colonies on wort gelatine delicate, commonly rosy above and below; odor slight but arsenical; conidiophore simple about  $15\mu$  by  $2\mu$  or none; conidia elliptical to lemon-shaped 3 to 6 by 2.5 to  $5\mu$ , “discontinuous” chain.

Biourge’s no. 393 (our no. 4733.103) upon Czapek’s solution agar produced a submerged slimy mycelium with no fruiting hyphae; upon wort agar slant a much wrinkled mass of mycelium, with coremium like structures rising 5 to 8 mm; later producing short pointed spore-bearing branches tipped by a mass of conidia 6 to 8 by 3 to  $4\mu$ .

The data available do not justify identifying it even to the genus.

519. *Scopulariopsis rubellus* Bainier. Bul. Soc. Mycol. France 23: 104; Pl. XII, fig. 6–11; 1907; not *S. rubellus* Bainier of Biourge’s monograph (no. 226).

Colonies on licorice sticks with aerial mycelium abundant, white, slowly becoming slightly rosy or even carmine, in age dark gray with a carmine tint, aerial hyphae about  $5.6\mu$  in diameter, variously arranged in ropes or fascicles which bear the conidial apparatus as perpendicular branches; conidial apparatus arising first as isolated, perpendicular sterigmata and groups of sterigmata irregularly distributed, later as penicillus-like branches with very short stalks and branching systems sometimes fairly simple and regular more often complex and consisting of several superposed series terminating in sterigmata with conidial chains; sterigmata described and figured as varying in length up to  $28\mu$ ; conidia very irregular, rarely spherical, 8.4 to 11.2 by  $5.5\mu$ , some of them truncate at the base, and others terminate in a point.

Described as a cause of decay in mushrooms. We can find no justification for Biourge’s (Monogr., p. 221) identification of this species with *P. amethystinum* Wehmer from examination of no. 4733.2a received from him under this name. No. 4826C2 received from Holden, Nottingham, England, satisfies Bainier’s description fairly well.

520. *Scopulariopsis rufulus* Bainier, Bul. Soc. Mycol. France 23: 105; Pl. XII, fig. 1–5; 1907.

Synonyms: *P. (Scopulariopsis) rufulum* (Bainier) Biourge. Monogr. La Cellule 33: fasc. 1, pp. 220–221; Col. Pl. VI, Cart. 17; Pl. X, fig. 55, 1923; fide Biourge, *Torula rufescens* Fresenius, t. XI, 11 to 17.

Colonies on licorice sticks at first white floccose (une masse), later approaching pale coffee color (café au lait); conidial apparatus arising



either as single sterigmata each with a chain of conidia and borne directly but irregularly upon and perpendicular to creeping or ascending hyphae or short stalked penicillus-like forms with symmetrical branching and forms in which sterigmata and metula-like branches are variously intermingled; sterigmata usually long, up to 15 times as long as broad, swollen toward the base and tapering gradually toward the conidia-bearing apex and often slightly sinuous; conidia average  $5.6$  to  $7\mu$ , with a truncate base and figured as somewhat angular at first "oval then more or less spherical," with a central more refringent vacuole or granule.

*S. rufulus* was found upon overripe cheese, and upon decaying mushrooms.

Culture no. 4640.466 labeled *S. rugulus* as received from the Bainier collection may have been the type of this species, but did not fit the description as well as no. 4500.382, labeled *Oospora crustacea*, contributed by Dr. Schmitter as obtained from the lungs of a bird by Dr. Moquet in Paris, or as no. 2731 received from Miss Dale in 1912. Miss Dale's culture (no. 2731) was redder than the usual types of *S. brevicaulis*, produced conidia almost globose  $5$  to  $7\mu$ , more or less tuberculate. The contrasting color was the most prominent mark of separation from the other species of Scopulariopsis in her collection. Biourge reported the conidia as  $5$  to  $9.5$  by  $2.5$  to  $9\mu$  and smooth; his no. 17 (our no. 4733.112.1) is noted as separated from *P. brevicaulis* by its wrinkled and buckled colony instead of *plane*. No. 4733.112.1 grew in Czapek's solution agar as one of the deep chocolate brown members of this series.

521. *P. sacculum* Dale. In Biourge Monogr. La Cellule 33: fasc. 1, p. 323; Col. Pl. XIII, Cart. 360; Pl. XXIII, fig. 134. 1923.

See Dale, Elizabeth, Ann. Mycol. 24; no. 1-2, p. 137, 1926, for proposal of the name from a description in Ann. Mycol. XII; p. 52, 1914.

The following is all of the descriptive information given in Miss Dale's 1914 paper; "10 *Penicillium* ? sp. (D6). Forms a flat thin culture on gelatine which is not liquefied, at first white then gray brown to ashy gray, with reverse uncolored; conidiophores short, unseptate, thin, erect, arising from creeping, septate, rather thick vegetative mycelium, sometimes branching irregularly but branches not cut off by septa; conidiophores cells may or may not be cut off by septa; they are much dilated and bear short chains of spiny thick walled conidia which are dark brown when ripe. Figures 99, 100."

Biourge cites only the name, and gives his carton and figures. Dale in

1926, proposes the name as new (cited by Biourge in 1923), and cites the figure and discussion given in 1914 in lieu of diagnosis.

Following our own records Miss Dale's D6 (Thom 2696) has been lost but was carefully studied and reported to Miss Dale as provisionally *Scopulariopsis repens* Bainier. Subsequent data, including the figures of Biourge confirm the assignment of the form to *Scopulariopsis* at least. Unless new material should be found to verify this species it should be dropped.

522. *Stysanus stemonites* Persoon? In Biourge, Monogr. La Cellule 33: fasc. 1, p. 216; Col. Pl. VI, Cart. 166; Pl. IX, fig. 51. 1923.

Upon all gelatine media (for all gelatine has at least a trace of arsenic) he obtained the arsenical odor of the *Anomala* series. Upon this and other grounds he transferred to *Penicillium* the species assigned by previous authors to *Stysanus*, making them *Penicillium*-section *Anomala*. We have cultivated *Stysanus stemonitis* from time to time over a period of twenty-five years and simply do not agree with Biourge upon this point. Biourge's no. 166 (our no. 4733.115) is apparently the organism commonly recognized as *Stysanus*.

523. *Scopulariopsis veneriei* Greco (N.V.). In Origine des tumeurs et observations de mycoses, p. 709-721, p. 823, pl. XXI and XXVIII, figs. 437-442, Buenos Aires. 1916.

Cultures on potato, carrot, glucose peptone agar and maltose peptone agar, this strain produced colonies, slightly yellowish grayish-chestnut, yellowish-chestnut to chestnut with yellowish radiate spots, spreading, with whitish gray ropes at the center and in streaks (striae), border whitish gray; mycelium abundant, septate, 2 to 5 $\mu$  diameter; with granulations in protoplasm 0.5 to 1 to 4 $\mu$ , rounded or rod-like, refringent, deeply yellowish; with piriform or slightly rounded swellings in older hyphae; conidiophores short, 10 to 20 $\mu$ , in length, arranged on main hyphae so as to give a verticillate appearance; fruiting mass in clusters, powdery, yellowish gray-chestnut; sterigmata terminal slightly conical or blunt; conidia in clusters, 5 to 8 by 4 $\mu$  with outer wall much thicker than wall of sterigma, globose but generally oval or lemon-shaped, with refractive drops on surface which are yellowish, 0.5 to 1 $\mu$ , giving a rugulose appearance, on crushing separating from wall of conidium and becoming larger, round or quadrangular.

The specific name was derived from the localization of this type of granuloma. Greco reports that Donovan (1905) described its cause as

an enormous bacillus 2 by 1 to 1.5 $\mu$ . Siebert and Flu refer to the cause as yeast forms; others suggest to protozoa; Greco in his cases reports the agent of this same clinical picture of venereal granuloma as a fungus and in two cases isolated the fungus described here. No identification is possible from Greco's description although the data given suggest that it was a *Scopulariopsis*.

524. *Acaulium violaceum* Sopp. Monogr., p. 56-60, Taf. IV and VIII, figs. 70-74. 1912.

Colonies violet-gray, with mycelium submerged at first, later producing aerial fruiting areas which develop aerial filaments and finally felted masses, resembling typical *P. brevicaulis*, on bread, however, the growth is luxuriant, gray-violet in color and showing as abundant corium formation, suggesting *Stysanus*; hyphae partly delicate, partly coarse, vesiculose or chlamydospore-like, with frequent formation of yeast-like cells or whole hyphae breaking into oidia; conidiophores irregular, often in bundles or ropes, or wanting, commonly *Penicillium*-like in young cultures and suppressed entirely in old cultures, or represented by feather-like bundles of hyphae fringed with conidial fructifications; sterigmata long, crooked (snake-like), sessile or stalked, recorded as occasionally suppressed in old cultures so that the conidia are produced directly on the mycelium; conidia violet-brown, large, oval, thin walled, smooth, up to 12 by 15 by 9 to 10 $\mu$ , pointed at one end, broad at the other; perithecia not found.

Species discovered upon bird bone in garden earth.

## CHAPTER XXIV

### PAECILOMYCES

534. *Paecilomyces* Bainier. In Mycothèque de l'École de Pharmacie, XI.

Bul. Soc. Myc. France 23, p. 26, plate 7, 1907. Spicaria of various authors: Corda?

*Corollium* Sopp. *Eidamia* of Horne and Williamson not Lindau. Type species *P. varioti* Bainier.

Bainier's generic description translated and emended: Genus related to *Penicillium* and *Aspergillus* distinguished by sterigmata short-tubular or more or less enlarged, tapering into long conidium-bearing tubes mostly curved or bent slightly away from the axes of the main sterigmatic cells; sterigmata variously arranged, partly in verticils and branching systems suggesting *Penicillium*, partly irregularly arranged upon short branchlets, partly arising singly along the fertile hyphae; conidia in chains, ellipitical, never green (fig. 96, 97).

Thom described a member of this genus as *P. divaricatum* and accumulated a large series of closely related strains. Bainier and Thom in their descriptions failed to discuss the accessory spore-like cells found mostly on young submerged branchlets or on branches close to the substratum in our species (no. 34), apparently more generally distributed upon other strains.

Horne and Williamson (whose cultures we have directly from them) working with an organism of this group (Annals of Botany 37: no. 147, p. 429, 1923) describe as macrospores, these large cells which are variously borne but usually solitary upon branchlets (or (?) metulae) which are either solitary or variously aggregated upon the fertile hyphae. They regard these "macrospores" as fixing the generic allocation of the species and transfer it to *Eidamia* Lindau. (See Die Pilze Deutschlands, Oesterreichs und der Schwiez., VIII Abt., Fungi Imperfecti in Rabenhorst's Kryptogamen flora, 2 aufl, Band I, 1907).

Lindau in describing the Genus *Eidamia* with *E. acremonioides* (Harz) Lindau (Syn. *Monosporium acremonioides* Harz—*Papulaspora aspergilliformis* Eidam) as its type species, gives the following significant characters: Hyphae branched septate, uncolored; conidiophores erect, branched, septate, narrowed toward the apex then forming a

globose vesicle; sterigmata radially arranged, covering the vesicle, acute pointed; conidia in chains hyaline; producing accessory fruiting masses as bulbils terminal on branches and on branches homologous to the conidium bearing branches producing one-celled ovate and yellow brown chlamydospores.

Lindau's generic characterization of *Eidamia* would exclude the *Paecilomyces* type of organism unless the revisers of the genus are prepared to show that *E. acremoniioides* (Harz) Lindau, the type species,

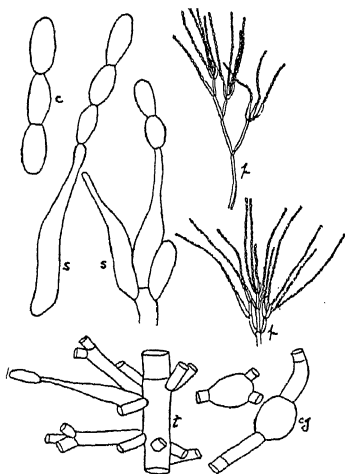


FIG. 96. *Paecilomyces varioti* Bainier: Thom's figure, 1910, showing, c, conidia; cg, germinating conidia; p, sketches of penicilli; t, section of a trailing hyphae bearing sterigmata and various grades of penicillus-bearing branchlets.

belonged in a series with Bainier's and Thom's strains, and had been incorrectly interpreted by Harz and by Lindau as well as by Eidam. Horne and Williamson include a common organism generally recognized as *Trichoderma* in their conception of *Eidamia* (under the name *E. viridescens* H. and W.) and place their *Trichoderma* next to *E. catenulata* H. and W., which is a *Paecilomyces*, and which in no way resembles the *Trichoderma* in question unless the common production of questionable cells with possibly superficial resemblance, designated by them macrospores, is to over-shadow all other characters in classification. Their organisms *E. catenulata* (our no. 4734a) and *E. viridescens* (our no. 4734c) have very little in common. Kita and Wai (report by Wai)

working at Kyoto isolated another member of this same series (our no. 4853) which produces these "macrospores" even more abundantly than the English strain. They prepared and sent us a tentative description, with detail drawings and photographs for their organism as a new species of *Penicillium* which is thus far unpublished. There appears to us little ground for considering these species to belong to *Eidamia*.



FIG. 97. *Paecilomyces varioti*: Diagrammatic radial section of the margin of a growing colony; showing loose submerged hyphae, the dense mycelial layer at the surface, aerial trailing hyphae and ropes of hyphae bearing penicilli.

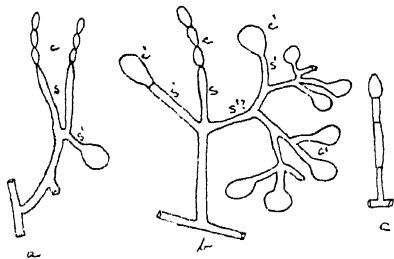


FIG. 98. *Paecilomyces*, *a*, *b*, Kita and Wai's species: Reproduction of a sketch by the describers which shows their interpretation of the so-called "macrospores" of Horne and Williamson as transformed units homologous to sterigmata and conidia; *c*, specialized branch and terminal cell of *Spicaria violacea* Gilman and Abbott which probably represents the same type of structure.

After Horne and Williamson's paper and cultures were received we restudied a series of forms including our own type strain no. 34, Kita and Wai's organism and several others. The specialized cells certainly exist. Development in the strains studied varies from the occasional isolated slender stalk and terminal cell of no. 34 to the complex clusters of Horne and Williamson and the clusters of Kita and Wai

(fig. 98) showing sterigmata with chains of conidia as branches homologous to those bearing "macrospores."

A type culture from twigs of *Salix* was described with the establishment of the genus by Bainier as *P. varioti*.

Since Bainier had but one strain, probably correctly represented in our no. 4640.436 received in the Bainier collection, separation of generic from specific characters was indefinite. This strain is so nearly identical with Thom's *P. divaricatum* that Thom's characterization of the species is given in slightly emended form.

Gilman and Abbott transfer all of these species to *Spicaria* and re-describe them there. Their culture of *S. elegans* when examined was clearly one of this group. The whole question of the generic diagnosis of *Spicaria* remains open. No attempt has been made here to review critically all organisms described as species of *Spicaria* and decide their relation to *Paecilomyces*. The usages already included show a wide disagreement in interpreting Corda's genus.

535. *Paecilomyces varioti* Bainier. Bul. Soc. Mycol. France 23: p. 26, Pl. VII, 1907. (Compare our figs. 94, 95.)

Synonym: *Penicillium divaricatum*, Thom, U. S. Dept. Agr. Bur. Anim. Ind. Bul. 118, pp. 72-73, fig. 29, 1910, and *Spicaria divaricata* (Thom), Gilman and Abbott, Iowa State Coll. Jour. Sci. 1: no. 3, p. 301; 1927.

Cultivated in gelatin or bean agar, yellowish brown (avellaneous), never green, broadly spreading in the substratum; superficial growth consisting mostly of trailing fertile hyphae, becoming powdery in appearance when mature; reverse of colony not discolored; fertile hyphae septate, usually short, mostly creeping; conidial fructifications either terminal or on short branches of creeping or partially erect hyphae, consisting of separate sterigmatic cells, of verticils, or of series of verticils of branchlets and sterigmata irregularly distributed along the fertile hyphae; sterigmata 15 to 20 by  $3\mu$ , with long acuminate tubes usually bent from the axis of the cell and broadly divergent at the apices, bearing long chains of conidia; conidia elliptical or fusiform, 5 to 7 by 2.5 to  $3\mu$ , yellowish to brownish, swelling in germination to  $10\mu$  and producing 2 or more tubes; does not liquefy gelatin; litmus reaction alkaline.

Unmistakable when once seen in culture. Found in a mucilage bottle, Storrs, Conn., 1904. Later contributed by Prof. G. F. Atkinson from North Carolina.

A large number of cultures with the characters of *Paecilomyces* have been seen. These vary in shade of color of colony, color in the substratum; in floccosity; in the abundance and distribution of the so-called macrospores, in the arrangement of the sterigmata and the size of the conidia. No one at present knows this group well enough to establish sound lines of relationship among them. Our collection and records show organisms from Japan, from Manchuria, from Europe, from South Africa, and from many parts of the United States. Viewed in conjunction with the descriptions and figures found in the literature, this structural type is seen to be cosmopolitan and is found described under the generic names *Corollium*, *Spicaria*, *Penicillium*, *Paecilomyces*, *Eidamia*, and *Byssochlamys*—perhaps more.

For purposes of analysis the following table based upon measurements of conidia begins with the largest conidia and includes the measurements given for described species which we have recognized or tentatively assigned here interspersed with measurements of conidia from strains in our collection.

Conidia 9 to 10 by 3 to 4.....	<i>Corollium dermatophagum</i> Sopp.
Conidia 6.5 to 10 by 5 to 6.....	<i>Spicaria fimetaria</i> Moesz.
Conidia 6 to 8 by 3.5 to 4.....	<i>P. repandum</i> Bainier and Sartory.
Conidia 5 to 8 by 2 to 3.....	4725.531 Hansford—Jamaica.
Conidia 5 to 7 by 2 to 4 $\mu$ .....	<i>P. Burci</i> Pollacci.
Conidia 6 by 3.....	<i>Paecilomyces varioti</i> Bainier.
Conidia 5 to 6 by 3 to 3.5.....	4777.11 from Pribram.
Conidia 4.2 to 6 by 3 to 4 $\mu$ .....	<i>P. flavum</i> El. & Em. Marchal.
Conidia varying 2.4 to 4.5 or 6.4 by 3.6; also 14 by 5.8.....	<i>P. aureo cinnamomeum</i> Biourge.
Conidia 4 to 7 by 2 to 3.5.....	Type of <i>Eidamia catenulata</i> Horn & Wil- liamson 4734a.
Conidia 4 to 6 by 2 to 3.....	4010.3 Turesson—Seattle.
Conidia 4 to 5 by 2, 5 to 7 by 3 to 5; 4854b.....	Kita's strain.
Conidia 4 to 5 by 2 to 3.....	4282.31.
	4777.27, <i>Byssochlamys nivea</i> from Pribram.
	<i>P. mandschuricum</i> Saito Type of no. 4279.
	<i>P. arenarium</i> S. & M.

Our type culture was found in an empty bottle of library paste.

Members of the *Paecilomyces* series have been found in the most diverse environments in nature. Turesson isolated a strain from human feces. Segal reported one as isolated from a guinea pig inoculated with the virus of typhus fever. We have had cultures from licorice root, from a hen's egg, from nut margarine, from soy products in China,



from bread in Arabia and from a quinine solution. Great masses of mycelium of this species were found deep in a pile of cabbage waste at a sauerkraut factory in Ohio where the temperature had reached 132°F. (about 55°C.). No determinations as to actual heat tolerance were made, however. Kita and Wai found their organism upon rotting boards in a cellar. Several strains were received from the Forest Products Laboratory of this Department at Madison, Wisconsin, as isolated from wood in various stages of discoloration or other injury from fungi. Other strains were contributed by Putterill at Cape Town, Van der Bijl at Durban, Natal. South Africa, C. G. Hansford in Jamaica, from Pribram in Vienna.

*Paecilomyces* may thus be regarded as a group of cosmopolitan saprophytes active over a wide range of temperature and upon the most varied substrata.

*Paecilomyces erectus* Demelius (see Chapter XX) as a green species which we have not seen but on the basis of the description given, we have placed it in the miscellaneous section of Biverticillata. *Penicillium divaricatum* (our no. 4777.10) from Pribram was a strain of *Scopulariopsis*.

The following species are included in the *Paecilomyces* group either from known cultures or by our interpretation of descriptions. In the lack of evidence as to real relationship, the names are arranged alphabetically to the species—disregarding the generic initial. Changes in generic nomenclature are only proposed in certain species known to us in culture.

536. *Penicillium arenarium* Shaposhnikov and Manteifel. Trans. Sci. Chem.-pharmaceut. Inst., Moscow 5: 1-64, figs. 1923. In Russian.

A laboratory contaminant growing at 40°C. under reduced oxygen pressure to which the name *P. arenarium* was given on account of its sandy color.

In artificial media as undiluted wort, glucose with 3 to 5 per cent of peptone and media containing a total nitrogen up to 0.2 per cent in contrast with the usual 0.05 to 0.08 per cent, mycelium white, 15 $\mu$  in diameter, twining; surface of colony olive sandy or sometimes greenish, even rose, light yellow, dark olive green to blackish, dark blue green in depressions between buckled areas (? bluish in effect C.T.); reverse characterless; conidiphores 150 by 5 to 15 $\mu$ , on short side branches of mycelium, having an indefinite tree-like branching; metulae when

present varying in number, and bearing 2 to 4 sterigmata, separated by septa from metulae, or entire structure non-septate (?—C. T.); sterigmata few, sometimes borne directly on the stalk,  $25\mu$  by 5 to  $6\mu$ , or even 75 to  $100\mu$  in length; conidia in extremely long chains, lemon-shaped with distinctly pointed ends, large, 10 to  $11\mu$  in diameter (largest dimension ?—C. T.), “gemmae” (of author) formed on ends of side branches, 14 to  $15\mu$  in diameter, thick-walled.

No perithecia were found. The optimum temperature was  $35^{\circ}$  to  $40^{\circ}\text{C}$ . In cultural experiments this organism tolerated 0.5 per cent acetic acid, 12 per cent of citric acid and 20 per cent of tannin according to the authors.

When the miscellaneous observations of the recorded cultural work are analysed and tabulated as above, the organism becomes a strain of the *Paecilomyces varioti* group.

Dr. S. A. Waksman of New Brunswick, New Jersey, was kind enough to translate this paper for us from the Russian.

The anomalous series of colors including bluish and greenish appear as indeterminate shades in the mycelium in various strains, not in the conidial areas which have not been green or blue in our experience with hundreds of cultures.

537. *Paecilomyces aureo-cinnamomeum* (Biourge) Thom. See *P. aureo-cinnamomeum* Biourge Monogr. La Cellule 33: fasc. 1, pp. 213–214; Col. Pl. V, Cart. 61; Pl. VIII, fig. 48. 1923.

Colonies on wort gelatine yellow to fulvous, cinnamon, reverse at first yellow then cinnamon brown; odor none; conidiophores 3 to  $4\mu$  in diameter; penicillus up to  $60\mu$  long, with walls smooth; branches irregularly produced; metulae 16 to 20 by 2 to  $4\mu$ , irregular in number; sterigmata single or paired, 13 to 30 by 1.5 to 2.5, producing long slender tubes bent from the median line; conidia ellipitical varying greatly in size 2.4 to  $4.5\mu$  by 2.4, to  $6.4\mu$  by 3.6, and also 14 by  $5.8\mu$ .

Biourge no. 61 (our no. 4733.10) is a member of the group called by Thom in 1910, *P. divaricatum*, by Bainier *Paecilomyces* and probably *Corollium* by Sopp, and by others *Spicaria*. Grown on Czapek's solution agar 4733.10 produced colonies thin, vinaceous to avellaneous, 100 to  $200\mu$  deep, with tangled ropes of hyphae bearing various groups of sterigmata 10 to  $20\mu$  or more in length of which the beaks 6 to  $10\mu$  in length were distinctly set at an angle to the axis of the cell; conidia 4 to 5 by 2 to  $3\mu$ , in very long tangled chains.

**538. *Paecilomyces Burci* (Pollacci) Thom.**

*P. burci*, Pollacci, Gino. Miceti del corpo umano e degli animali. Ist. Bot. d. R. Univ., Pavia II ser., 18:128-129. Tav. XXXI, figs. 4-6. 1921; also in Contributo allo studio dei granulomi sperimentali provocati dal *Penicillium burci* n. sp. by A. Campatelli, Pensiero med., Milano, 12:217-19, 1923.

Isolated in 1920 by Guiseppe Berti from an experimentally provoked nodule. In Sabouraud broth and the same broth with peptone and glucose at 18°C., colonies cottony in two to three days with delicate, short sterigmata bearing yellowish white, little conidia in chains, avel-laneous; mycelium, septate, creeping or ascending, 6 to 7 $\mu$  in diameter; conidiophores erect, septate hyaline, 50 to 110 $\mu$  in length, bearing small branches; conidia in chains globose, rarely ellipsoidal, pale fuliginous, 4 to 5 $\mu$  in diameter. Pollacci notes that at some stages this form resembles *Oospora* as described by Wallroth (fig. 4 of Tav. XXXI) and at other times forms terminal swellings which suggest early stages of *Acremoniella berti* Pollacci found associated with it.

A culture contributed by Pollacci and received from the Centraal-bureau (our 4876.25) presented conidia in chains, as well as fusiform and larger clavate, terminal forms. Pollacci's figure gives a mono-verticillate form with part of the conidia shown in chains as definitely elliptical, the others rather less definitely globose. Our notes follow: Colonies in Czapek's solution agar, broadly spreading forming a loose bristly network of hyphae and ropes of hyphae (funiculose) making a felt 200 to 400 $\mu$  deep, white with more or less yellow in areas, and with yellowish drops or their residues, reverse in yellow orange shades; specialized conidiophores scarcely found; penicilli consisting of single sterigmata or small short-stalked clusters or verticils of sterigmata each with its conidial chain, scattered along the separate hyphae or ropes of hyphae, in large numbers; sterigmata 10 to 12 $\mu$  with long tubes, set more or less at angles with the main axes of the sterigmata and divaricate when in verticils; conidia up to 6, 7 or even 8 $\mu$  long by 2.5 to 4 $\mu$  in diameter, almost colorless.

**539. *Corollium dermatophagum* Sopp. Monogr., p. 99-103, Taf. X, fig. 108, Taf. XXIII, fig. 45. 1912.**

Colonies smooth, forming at first a thin greenish veil-like covering of the substratum upon which conidial areas develop slowly, in old cultures becoming reddish or chocolate brown; vegetative hyphae are moderately coarse with fruiting hyphae somewhat coarser; conidio-

phores coarse, commonly with granular walls and arising perpendicularly from the coarser hyphae, branching variously to produce several types of conidia-bearing branches; branches bearing scattered clusters of sterigmata and terminating in large clusters of sterigmata which are always in verticils and figured by Sopp as larger and clustered at the base with long tapering diverging beak-like conidia-bearing tubes, the whole up to 50 to 60 $\mu$  long by 10 to 12 $\mu$  in largest diameter; conidia clear yellowish brown, oval, smooth, 9 to 10 by 3 to 4 $\mu$ , persisting in very long chains; structures suggestive of perithecia are reported but without recognizable descriptions or figures.

Found first upon rotten leather and later common upon miscellaneous substrata in the laboratory; liquefies gelatine, upon colonies at first veil-like, suggestive of *P. aromaticum* (*P. roqueforti*) show yellowish brown or greenish yellow colors, later becoming dense and wrinkled, with reverse dirty green; optimum temperature for growth 38° to 40°C., maximum above 45°C. Sopp reported extensive experimentation upon different substrata in which it produced (Sopp, p. 102) malic and citric acids, catalase, chymosin, pepsin, and diastase, as well as small amounts of alcohol, ether and other aromatic substances.

539a. *P. elegans* Corda. Icon. II, Table XI, fig. 74, p. 18. 1838.

Synonym. *Spicaria elegans* Harz.

A delicate white verticillium-like form figured with 2 to several times verticillate diverging branching systems producing long curving chains of elliptical conidia borne upon diverging long-taper pointed sterigmata few in the verticil.

Many workers place all of the genus *Spicaria* here as for example, Gilman and Abbott.

540. *Spicaria fimetaria* Moesz. Botanikai Közlemenyek 19:58, fig. 9 (p. 59), 1921.

Latin diagnosis translated (C. T.): Effused, rosy, powdery; sterile hyphae creeping, branched, septate, about 5 $\mu$  in diameter; fertile hyphae suberect, septate, irregularly branched; branches frequently bearing dichotomous branchlets; ultimate branchlets (sterigmata—C. T.) with apex acute producing conidia ellipsoid, hyaline, smooth 6.5 to 10 by 5 to 6 $\mu$  and in long chains.

Habitat: in horse dung in the field at Teteny near Budapest, collected by F. Hellendonner.

Moesz's figures suggest *P. divaricatum* in the general branching of the conidiophore, the general size, arrangement, and the long curved beaks

of the sterigmata. They fail to show the real manner of conidium formation, however. The measurements of the conidia are somewhat what larger than the usual form of *P. divaricatum*.

541. *Penicillium flavum* El. and Em. Marchal. Bul. Soc. Roy. Bot. Belgique, T. 54, (Ser. 2 T. IV), p. 129. 1921.

Colonies flavous rarely in age verging toward ochraceous; hyphae guttulate, septate, densely felted, frequently anastomosing, at times ascending as floccose fascicles (ropes ?); conidiophores erect, simple or ("coalitis") funiculose, septate, 145 to 190 by 4 to 5 $\mu$ , branched above in 3 to 4 series; sterigmata frequently in 3's, scarcely attenuate at the apex, 13 to 19 by 2.3 to 3 $\mu$ ; conidia ovoid, hyaline, smooth, frequently 1-several guttulate, 4.2 to 6 by 3 to 4 $\mu$ .

Species found upon fruit, apples, pears, cherries, etc., in Belgium.

The Marchals add the note that a form grown upon pear branches showed conidia slightly smaller 3.8 to 5.7 by 2.3 to 3 $\mu$ .

542. *Paecilomyces mandshuricum* (Saito) Thom n. comb.

*P. mandshuricum* Saito. In South Manchuria Railway Company, Central Laboratory, Report no. 6, p. 11-12, December 20, 1921. In Japanese.

Synonym: *Paecilomyces* sp. indet.

Appearing on wine yeast generally as a mass of olive green or olive yellow spores, occasionally as aerial hyphae having a thread-like appearance, in artificial media developing a thick membrane which bears a large number of spherical and smooth chlamydospores; stalks very tiny, 170 to 180 $\mu$ , irregular; spore bearing branches one to three, irregular, each bearing a chain of spores; conidium smooth, elliptical to ovate, generally 6.5 by 4.5 $\mu$ , some as great as 10 $\mu$  (germinating?—C. T.); optimum temperature 36°C; gradually liquefies gelatine and starch media; secretes as enzymes maltase, protease, peroxidase and catalase.

According to Saito this fungus bears a striking resemblance to *P. olivaceum*, but may be separated by the difference in their optimum temperatures, the latter preferring 23° to 25°C. Our no. 4279 received as *P. mandshuricum* in 1918 from Saito is a strain near *Paecilomyces varioti* Bainier, as discussed in this paper.

543. *P. repandum* Bainier and Sartory, Bul. Soc. Mycol. France 29: pp. 367.

Colonies on licorice sticks, fruiting in about six days, at first pale yellow then in deeper shades to dark orange yellow (C.d.C. 178); peni-

cillus rarely terminal, usually borne upon irregularly produced short branches of creeping or ascending hyphae enlarging slightly from base to apex and bearing single verticils of diverging sterigmata with their conidial chains, or more or less biverticillate by the development of part or all of the sterigmata into secondary branches with their verticils of sterigmata; sterigmata 5 to 6 or more in the verticil, 15 to 20 by 7 to  $9\mu$ ; conidia variable in size mostly 6 to 8 by 3.5 to  $4\mu$ , smooth, in chains, yellowish (C.d.C. 178D), swelling to double size in germination and sending out several tubes.

We have not seen this species but the description would place it among the Paecilomyces strains rather than the strictly penicillate species.

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## CHAPTER XXV

### UNDETERMINABLE PENICILLIA

Species have been assigned to sections of the whole group if diagnosis, figures or notes furnish even a possible justification for it. References to such species in the literature gain some value from allocation to membership in groups of related forms. In spite of diligent search, we have been unable to find justification for grouping many forms described by various authors from Link even to recent workers. Such species are, therefore, given in this chapter in alphabetical order to specific names irrespective of the generic name used.

550. *P. aeruginenum* Sopp. Monogr., pp. 145-147, Taf. XVI, fig. 115; Taf. XXII, fig. 11. 1912.

Colonies on meat-peptone-sugar-gelatin blue-green (aeruginous or?), (Niagara green), with a yellowish tinge becoming gray brown in age, developing large masses of conidia, with mycelium white becoming brownish in age and spreading very irregularly over the substratum; hyphae coarse; conidiophore comparatively slender, septate, rather long, forked with the development 4 or 5 branches with a common origin, each bearing verticils of metulae and sterigmata with ultimate verticils and sterigmata suggesting the *P. luteum* series; conidia 4 to 5 $\mu$ , globose, smooth, in long adherent chains; perithecia (or sclerotia—C. T.) green, once found and not described.

This species was found in earth, with a growth optimum at 20°, minimum at 5° and a maximum at 35°C. Colonies grew well on gelatine and agar, upon milk where the mycelium was reddish and produced a rosy velvety surface growth in urine, in broth, in wort, on potato, on bread, and rice. Conidia remained viable more than three years.

From the description his colonies were azonate, produced masses of conidia, branching of the conidiophore was described as triverticillate; red color was produced on milk, but not on other media; conidia were globose 4 to 5 $\mu$ .

550a. *P. (Acaulium) albo-nigrum* (?) Sopp. Appears in Biourge Monogr., Col. Pl. VI, no. 6. No other reference to the name is given hence it is probably an error.

551. *P. album* Preuss, *Linnaea* 24: 135. 1851.

Colonies effused white; conidiophores erect, simple unseptate; penicillus consisting of a 2 to 4 times verticillate system of branches, with part of the verticils incomplete (subverticillate); conidia ovate white.

Reported upon *Dictydium cernuum* in Hoyerswerda but without details enough to insure identification since many *Penicillia* usually green appear more or less white when grown upon decaying stems and leaves.

The name has been used by Epstein (Epstein, S. *Archiv f. Hyg.*, Bd. 45 (1902) Hft. 4, p. 360) for the Camembert cheese mold, *P. camemberti*, and by Eichelbaum (*Verhandl. Naturw. Ver. Hamburg* 1906, 3 Folge, XIV. p. 37) for a form found upon fallen wood in German East Africa, but no data as to the basis of the identification are given.

552. *P. album* Rivolta Paras. Veget., p. 452. 1873.

Morphology like *P. glaucum*.

553. *P. aspergilliforme* Bainier. *Bul. Soc. Mycol. France* 23: p. 14, Pl. IV, fig. 17-23. 1907.

Colonies upon licorice sticks floccose, green to dark or sordid green in age; mycelium forming a considerable aerial felt, with hyphae 2.8 to 5.6 $\mu$ ; conidiophores figured as short vertical branches of creeping hyphae, few septate, swelling at apex to claviform to flask-shaped, and producing single verticils of densely crowded sterigmata, or occasionally a branch and secondary head, from the figure the conidial chains probably formed a dense column; conidia globose, said to vary in size with the diameter of the hyphae hence 2.8 to 5.6 $\mu$ , recorded as swelling in germination and producing 3 germ tubes.

Species reported as found many times.

554. *P. aureo-flavescens* Biourge. *Assoc. des anciens élèves de l'école supérieure de brasserie de l'université*, no. 3, p. 32, Louvain, 1920.

Biourge places here a strain of the "rotondispores" of Dierckx with smooth spores under the above name without description.

555. *P. barbae* Castellani (1907). Cited in *Manual of Tropical Medicine*, Aldo Castellani and A. J. Chalmers, all editions, as found "by us growing on beard of natives of equatorial Africa and Ceylon." Neither any real description of this parasite nor the parasite itself have been seen by us.



555a. *P. benzoicum* Kossowicz. Ztschr. landw. Versuchswesen, Oesterr. 14; 69-70. 1911.

In a factory using gum benzoïn as a preservative, Kossowicz found a sample of gum with a peculiar phenolic odor. The gum was moldy and the species was readily isolated. Colonies upon potato in test tubes were during the first week dense white, becoming pale brownish in age; upon mineral nutrient solutions with citric acid growth was abundant giving persistently pure white colonies after many months. The conidiophores were 200 to 300 $\mu$  long, with side branches irregularly produced and again variously branched with every branchlet bearing 2 to 3 sterigmata 10 to 14 by 2 to 4 $\mu$  producing conidia oval or ellipsoid 4 to 6 by 3 $\mu$ ; upon sterilized gum benzoïn, perithecia were occasionally produced as yellow bodies ovate up to 2 mm. by 1 to 1.5 mm. The asci were ellipsoid with 4 to 8 spores, 4 to 7 by 3 to 4 $\mu$ . Temperature optimum was 20 to 25°C.

555b. *P. Bombycis* in Sopp. Videnskapselskapets Skr. f. Mat.-Nat. Kl. Kristiania, 1911, no. 2, p. 26. 1912.

Colonies upon gelatine media vigorous, yellowish white, later chrome-yellow to greenish, long remaining sterile, then suddenly putting forth abundant globules of fluid followed by development of perithecia as "truffle-like" bodies about 0.2 mm. in diameter, at first gray, then greenish, later olive green, with red areas showing (?); ascospores produced after a resting period of three weeks.

The species was found parasitic on the feet of living caterpillars, where it produced conidia globose 2 to 5 $\mu$  in diameter upon pear shaped sterigmata. No other descriptive data were given except that conidia characterized the parasitic phase and perithecia the saprophytic phase of growth. Full description was promised in his monograph but is not found there unless his description of *P. parasiticum* which suggests this description covers the same organism. No one has since identified this species.

556. *P. brachiatum* Ellis and Morgan. The label of this specimen reads, "Ohio, on ash bark, Morgan, 750."

Accompanying description in Morgan's handwriting states hyphae 600 to 700 by 6 to 8 $\mu$ , distinctly septate, with short, brush-like, opposite branches, bearing conidia in a loose head; conidia acutely elliptical, 4 to 5 by 1.5 $\mu$ .

Examination of the material shows a strain of *Clonostachys*.

557. *P. caespitosum* E. and E. The label of the herbarium specimen reads, "Maine, Harvey no. 58."

The description in Ellis' handwriting accompanying the specimen reads "fertile hyphae fasciculate forming small greenish-yellow tufts about  $\frac{2}{3}$  mm. high and broad, contracted below, spreading above, conidiophores erect, slender,  $2.5$  to  $3\mu$  in diameter, with short, erect branches at the apex bearing chains of conidia, globose,  $1.5\mu$  in diameter and forming a compact head  $20\mu$  in diameter."

Examination shows colonies yellowish or reddish, in tufts on wood; conidiophore septate,  $3\mu$  in diam.; metulae in one verticil producing one group of sterigmata typical of the *P. luteum-purpurogenum* group; sterigmata 9 to  $10\mu$  by  $2\mu$ ; conidia, rough, 2 to  $2.5$  by  $2\mu$ , as in *P. rugulosum*.

557a. *P. candidum*. Link Obs. 1809, p. 17; also, Morini, F. Sulla forma ascofora del *Penicillium candidum* Link in Malpighia 1888, p. 224. •

Synonyms: *Rhodocephalus* Corda—fide Bonorden Hdbch., p. 75; *P. camemberti* var *Rogeri* Thom according to Epstein and to Mazé; *P. epsteinii* Lindau based upon *P. camemberti* var. *Rogeri* Thom.

The description given by Link is not sufficient to identify this form.

Link gave: "b. floccis ramosis. *P. candidum* caespitibus rotundis et effusis, floccis candidis, capitulis sporidiisque concoloribus. In fungis herbisque putridis caespites majores minoresque format, floccis vix lin, altis."

The ascosporic form discussed by Morini has not since been reported. The name *P. candidum* has been applied by some workers to the white *Penicillium* used by Roger and his followers upon Camembert and Brie cheeses but the name appears only to signify that their organism is a *Penicillium* without reference to its identity with Link's material.

There are certain white species but it seems best to place them in their proper allocation in different groups under other names without trying to claim identity with Link's species. The occurrence of colorless colonies upon certain natural substrata, sticks, leaves, etc., is a common observation but when cultures are made from these colonies various green species develop thus indicating that the color of the original colony was due to the nutritive deficiencies of the substratum.

*P. candidum* in Saccardo's *Fungi italici* no. 892 was found upon a moldy bulb and showed conidia 3 by  $2\mu$ . *P. candidum* is cited by Bonorden as the basis of the genus *Rhodocephalus* Corda.

557b. *P. candidum* var. *coremioides* Sacc. Syll. IV, p. 80.

Saccardo transferred to *Penicillium* under this varietal name, *Coremium candidum* Nees.

No organism has been identified by us under this name although Prof. J. E. Cunningham has verbally described a white coremiform species with small elliptical spores as a cause of rot in certain vegetables in New Zealand.

We have seen a slide at the Cryptogamic Laboratory at Harvard labeled in Dr. Farlow's handwriting *P. candidum* with coremiform bundles and elliptical conidia but we have not seen it in culture.

558. *P. candidum* var. *subcandidum* Peck. N. Y. State Mus. Rept. 47, p. 22. 1894.

This variety was reported on *Agaricus campestris* in a greenhouse at Ithaca, N. Y., and is characterized "fertile hyphae irregularly branched above, the color at first white then whitish or cinereous."

Peck's variety is as unidentifiable as the species.

560. *P. capitatum* Ellis and Galloway. Herbarium material in Ellis Collection of the New York Botanical Garden. The label reads "on chinch bugs. Nov. 1892."

When examined in 1918, no *Penicillium* could be found.

561. *P. chrysomphalum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 323. 1923.

This is mentioned only as a beautiful fungus which he was too tired to finish hence left out of the book (naturally a nomen nudum.—C. T.)

563. *P. cinnabarinum* Fuckel. Symbolae Myc. Beitr. Kenntnis der rhein. Pilze. Zweiter Nachtrag, p. 79. Wiesbaden. 1873.

Reprinted from Jahrbucher des Nassauischen Vereins für Naturkunde 27 and 28.

No. 2589 of F. rh. ed. I and no. 404 in v. Höhnelt collection at Farlow Herbarium.

Colony in dense masses, effuse, cinnabarinus, with fruiting hyphae erect, branched, conidia in chains at apices, generally ovate, apiculate at both ends. On pigeon dung in the spring, common.

Microscopic examination of Fuckel's specimen in the v. Höhnelt Collection shows powdery, not ropy, cinnabarinus patches on the dung with the material in mass in a potassium hydroxide mount dirty cinna-

mon. Conidia thick-walled, some cinnamon color having a center rosy or deeper cinnamon, smooth or faintly rough, more apiculate at one end than the other, sometimes plainly truncate at one end in regard to outer face of wall, 7.5 to 13 by 4 to 7  $\mu$  in diam.

Not *Acrostagmus cinnabarinus* Corda, but related to *P. insigne* Bainier.

564. *P. citreo-lateritium* Biourge. Assoc. des anciens élèves de l'École Supérieure de Brasserie de l'Université, no. 3, p. 32, Louvain, 1920.

Biourge places a strain of the "ovalisporae" of Dierckx under the name citreo-lateritium without description.

565. *P. congolense* Dierckx. Soc. Scientifique Bruxelles 25, p. 87. 1901  
"*P. congolense*. Spores ovales de 2 by 3  $\mu$ .—Sterigm. 1-4 de 10 by 3  $\mu$ .—Fructif. 60  $\mu$ .—Formes massives. Thalle mince, peu consistant, poudreux. Spores vert-olivâtre assez stable. Revers et milieu colores en brun-fonce.—Moins caracterisé."

Biourge Monogr., p. 167, summarizes the above and suggests a relationship to *P. griseo-fulvum* and that this form may belong in his group hemizonata, section inflata but apparently was unable to recognize the species. It may be discarded.

566. *P. coremioides* Sacc. 1886., cited in Biourge Monogr., p. 102.  
*P. roseum* var. *coremioides* Kickx in Sacc. Syll. IV, p. 83; cited by Saccardo from Kickx Flore Crypt. Flandres, Vol. 2, p. 306, 1867.

Comparison of the page in Kickx shows that he did not use the name *P. coremioides* at all, nor "var. *coremioides*" but cited certain rosy spored forms as *P. roseum* Link without data which would warrant Saccardo's varietal designation. While it is not impossible that all of these "rosy" spored forms belong together there is not enough information in Kickx to warrant basing a name upon his report.

567. *P. croceo-hyacinthinum* Biourge. Assoc. des anciens élèves de l'école supérieure de brasserie de l'universite, no. 3, p. 32, Louvain, 1920.

Biourge places a strain of the "rotondisporae" of Dierckx with smooth spores under the above name without description.

567a. *P. crustatum*.

A misspelling for *P. crustaceum* in an article by Castellani on haemorrhagic bronchitis.

568. *P. cupricum* Trabut. Bul. Soc. Bot. France 42: 451-455, 1895.

See also De Seynes, *ibid.*, p. 482-485, and Bul. Soc. Mycol. France 15: 21, 1899.

Trabut described a species with red conidia growing upon copper sulphate solutions. De Seynes and Gueguen showed that the color and structures reported by Trabut were merely the responses of a green species to the copper sulphate. What strain Trabut used is apparently undeterminable.

Biourge (1925) reports a strain of *P. cupricum* from Estienne et Musquin which tolerated 10 per cent sulphate of copper in solution and resisted contact for a year with crystals of copper sulphate (with 5 molecules of water). He added that many *Citromyces* are very tolerant of copper.

569. *Penicillium cyaneo-carmineum* Biourge. Assoc. des anciens élèves de l'école supérieure de brasserie de l'université, no. 3, p. 32, Louvain, 1920.

Biourge places under this name a strain of the "rotondispores" of Dierckx with smooth spores, but includes no further description.

570. *P. deformans* Sopp. Monogr., pp. 184-186; Taf. XXI, fig. 145 (196 in text); Taf. XXIII, fig. 32. 1912.

Colonies on meat-peptone-sugar-gelatine white, fibrous, clear blue becoming pale then progressively changing through shades of gray to mouse gray sprinkled with white spots (overgrowth?—C. T.), never blue on potato and rice which are mouse gray from the first; reverse whitish to reddish or reddish yellow; hyphae delicate, uneven, sinuous; odor none or not definite; conidiophores slender, irregular, short, branched, in Sopp's figures short branches from trailing or ascending hyphae 1-celled or not more than 2-celled, with terminal verticil of divergent metulae or a single verticil of sterigmata; metulae when present irregular, diverging, usually enlarged at the apex with *Citromyces*-like conidia production; sterigmata figured as short flask-shaped narrowing to sharp divergent points bearing diverging chains of conidia; conidia 4 to 5  $\mu$  in long axis rough in long chains, separated by connectives.

Perithecia (sclerotia? more probably) developed only on potato, large, in clumps, with surface tufted with reddish hyphae which bind them into clumps. Asci not reported.

Species found in earth in West Norway. Cultures grew best at 20° with minimum at 3° and maximum at 33° C., and grew well in the common media tested. Conidia remained viable more than three years.

No one except Sopp has reported this species, but we have seen cultures of *Aspergillus versicolor* in which head formation has been largely suppressed by the substratum used, in which the figure given by Sopp is quite characteristically reproduced.

571. "*P. dubiosum* Wehmer." Cited only by Doebelt, H., in Ann. Mykol. 7 (4): 315-338, 1909, in his discussion of *P. africanum*; no description or other reference has been found by us.

572. *P. epigaeum* B. and C. N. A. Fungi no. 677 in Grevillea 3: 111-112, March 1875; cited *P. epigaeum* B. and C. in Saccardo Sylloge 4: 82. 1886.

Colonies on earth, heavy, fulvous; conidiophores branched from the base, with branches cuneate upwards; conidia at first elliptical then globose 13 to 15 $\mu$  in diameter, showing a "connective."

Collected on earth in New England.

A guess only from this description would place this form among the Scopulariopsis group, but closer identification is not possible.

573. *P. fasciculatum* Sommerfelt. Suppl. Florae Lapponicae, p. 342. 1826.

Hyphae fasciculate erect, with apex trifid, with branches again branching and spore bearing; spores cinerascens.

Sommerfelt's species was formed upon "*Sclerotium durum*" on the stems of Rumex. He cited *P. glaucum*,  $\beta$ . *fasciculatum* Pers. Myc. 1 p. 412 as the same species. The name has been repeatedly cited but no ground for identification is offered by the original description unless we accept his own guess of identity with Persoon's fasciculate variety of *P. glaucum* which may have been the apple rot organism, *P. expansum*.

In describing this species, trichotomous branching of the conidiophore is strictly specified, the absence of coremia is noted but no other data are given to aid in identification except frequent occurrence upon *Sclerotium durum* which is the basis of Berkeley's use of the name (Berkeley, M. J., in Ann. Nat. Hist., Ser. 1, Vol. 1, p. 262, 1838, and British Fungi Fasc. 3, no. 210).

574. *P. fieberi* Corda. Prachtflora, pp. 19, 20; Taf. IX. 1839.

Colonies effused, broadly spreading, delicate, pale olivaceous to dark brown; conidiophores erect, colorless; conidia globose, verrucose, olivaceous, unequal in size, the terminal spore largest, in chains, at first in an ovate mass or head, then spreading loosely.

Species found upon an insect (*Pentatoma prasina*) in Prague in 1836.

This species has never been recognized since Corda's description, although reference to the occurrence of terminal conidia much larger than others in the chain is made several times by Biourge under the caption "the phenomenon of Corda" (see Chapter VI).

575. *P. flavo-fuscum* Biourge. Assoc. des anciens élèves de L'école supérieure de brasserie de L'université, no. 3, p. 32, Louvain, 1920.

Biourge places a strain of the "rotondispores" of Dierckx with smooth spores under the above name but without further description.

576. *P. glauco-ochraceum* Preuss. (G. T.). Linnaea 24, p. 135, 1851, in F. Hoyersw. no. 122.

Many *Penicillia* if grown upon decaying wood, might easily satisfy this description which indicates a conidiophore producing at its apex one branch, or two, making a verticil of 2 or 3 metulac, and closely aggregated verticils of sterigmata with divergent chains of globose, greenish conidia. No cultures were made.

Biourge repeats the name in his "list onomastique" and attributes it to Sopp.

577. *P. glaucum* Link. Obs., p. 17, 1809.

The original reads:

"a. Floccis simplicibus.

*P. glaucum*, caespitibus effusis, floccis albis, capitulis sporidiisque demum glaucis. In corporibus putridis frequens. Maxime affine *P. expanso*, et forte ipsius varietas nondum perfecta. Iconem v. fig. 24."

Since controversy as to the use of names has characterized most of the intervening century, the original is given as justification of the classification *entirely undeterminable*.

Brefeld in his classic "Untersuchungen" in 1874, using the name *P. glaucum*, figured his organism elaborately but in diagrammatic form accompanied by description of the production of sclerotia. These sclerotia after a period of rest produced perithecia (Brefeld p. 70-71)

with ascospores 5 to 6 by 4 to 4.5 $\mu$ , 2-valved as in the *Aspergilli*, each valve bearing 3 to 4 ridges which appeared granular by ordinary (?) magnification. On page 26 of his paper he reported the conidia as 2.5 $\mu$ , globose, smooth in mass yellow. In one figure, he showed coremia suggestive of *P. expansum*. The ascospores as described and figured are almost identical with those of *Aspergillus fischeri* Wehmer.

578. *P. glaucum* Link. in Sopp Monogr., pp. 140-141, quoted from his book *Über Käsevergarung*, Christiania 1905, I. Sauermilchkase, p. 68. 1912.

Colonies green with a yellow tinge, surface coarse and rough, dense mycelium, spreading slowly, without wrinkles; odor intensely moldy with a suggestion of naphtha, suggestive of rotten oranges, taste of mold in cheese bitter, obnoxious; grows best between 15° and 25°C., poorly above 25°C. and not above 30°C.; liquefies beer wort gelatine in five days at 15 C.; grows fairly well under partly anaerobic conditions where it produces white perithecia in 4 days without green conidial areas; dissolves casein.

Sopp reports *P. glaucum* as a typical food and fruit rot, as well as active in the soil, always present in the air and in the soil, especially tilled soil, although it may be absent or rare in forest soils. He regards it as occurring in a number of varieties rather than as a single uniform type. Upon page 78 he noted that the "Penicillium upon apples (always *P. glaucum*) under certain conditions forms coremia." This observation merely shows that he believed the apple rot organism belonged with the one he regarded as *P. glaucum* in his cultures but does not prove it since his discussions of his cultures do not indicate *P. expansum*. His report of perithecium formation definitely excludes the organism he described from the *P. expansum* relationship so far as we know it.

578a. *P. glaucum* f. *epimyces* Sacc. Myc. Ven. no. 1060.

This is listed in Michelia 1, p. 109 and p. 593 but apparently refers to *exsiccati* only. No description has been found.

579. *P. glaucum* var. *fetidum* Sopp. Monogr., pp. 141-143, Taf. XVI, fig. 110; Taf. XXII, fig. 1 and 2. 1912.

Colonies on meat-peptone-sugar-gelatine blue-green with a yellow tinge, surface rough (= tufted?), zonate, with mycelium white above reddish yellow (orange) in reverse; hyphae coarse; odor intense, sugges-



tive of a mixture of mold, naptha and rotten orange odors; conidiophores long, branching from the upper 2 to 4 nodes and producing verticils of metulae and sterigmata at different levels thus forming a long penicillus with complex branching; sterigmata comparatively long, cylindrical in verticils of 5 to 10 or more; conidia at first oval then globose smooth, blue-green, 5 by 5 to 6 $\mu$ ; perithecia with long ripening period, oval asci and "spores with rings" produced sporadically (not described and not figured by Sopp).

This variety was found in cultures from air and earth; colonies grew between 3° and 33°C. with an optimum at about 18°C., grew well in milk, urine, broth, wort, potato, rice, bread, earth and sawdust; conidia survived in culture less than a year. This description taken with the colored figures in Plate XXII indicate a heavy zonate colony with a wide white border and reverse intensely yellow with strong odor and spores large 5 by 5 to 6 $\mu$ .

580. *P. glaucum* var. *pallidum* Sopp. Monogr., p. 145, Taf. XVI, fig. 109. 1912.

Species characterized by an offensive odor upon meat-peptone-sugar-gelatine replaced by an aromatic odor in rice cultures, which were only slightly yellowed; mycelium comparatively thin, white, becoming yellow and in some areas bluish with the developing conidia; reverse citrine rather reddish yellow (orange); conidiophores as figured showed less complex branching than variety "fötidium"; conidia 5 to 6 $\mu$  in diameter.

This blue-green variety was found in cultures from earth; it was in general a weaker form than the variety *fötidium* also described by Sopp.

581. *P. glaucum* var. *inodorum* Sopp. Monogr., p. 143, Taf. XXII, fig. 3. 1912.

Colonies yellow green; odor slight, somewhat rancid in milk cultures; conidia up to 7 $\mu$  in diameter.

Sopp added quantitative differences in catalase number, acidity produced, and rate of coagulation of milk as a basis for separating his variety.

582. *P. glaucum* Lk. var. *crustaceum* Fr. "63. Ch. Spegazzini. Fungi Guaranitici. (1885-1889). Speg. \*\*11. no. 381. Skin of citrus fruit. Guarpi." Farlow Herbarium.

This specimen in the Fungi Guaranitici is plainly mislabeled.

584. *P. glaucum* var. *epixylon* Thüm. in de Thümen. Mycotheca universalis. Gallia. Cent. XIX, Wien. 1881. Lyon in ligno putrido nudo Juglandis regiae Lin. Apr. 1880. leg. J. Therry.

As herbarium material of *Penicillia* can not be identified by present technique because of fragmentation, this variety must be discarded with the species, *P. glaucum*.

585. *P. gliocladioides* Preuss. Fungi Hoyerswerda no. 16. Linnaea 25: 729. 1852.

Colonies white, stalk erect, tripartite and penicillately branched above with short chains of globose, hyaline spores; upon fallen branches.

No identification is possible.

586. *P. gliocladioides* Spegazzini. Myc. Arg. V. in Anales Mus. Nac. Buenos Aires, Ser. 3, T. 13, p. 433, 1912; see Sacc. Syll. 22: p. 1277, no. 8039.

Not *P. gliocladioides* Preuss, q.v., hence the name would be untenable, if the organism should be rediscovered.

Described from dead leaves of *Coffea arabica*, as with superficial mycelium very delicate, creeping, arachnoid, with hyphae 2 to 3  $\mu$  in diameter, producing conidiophores 30 to 100  $\mu$  by 3  $\mu$ , erect or curved, septate, unbranched or bifid with apex producing 3 to 6 metulae, bearing sterigmata 5 to 10 by 1 to 1.5  $\mu$  in crowded verticils, with conidia 5 to 6 by 4  $\mu$  forming a gray cylindrical to clavate mass 100 to 120 by 20  $\mu$ .

This description from a mass of moldy leaves of the coffee tree would not identify the species if found in any other situation.

- 586a. *P. grande*, listed in Hallier's article on parasitology in Flora 51: 297, 1868; and in Zschr. f. Parasitenkunde I, 1868, as associated by Hallier with typhus. The species was not described. It is mentioned once more by Cavara and Mollica in Ann. Mycol. 5: 121-149, 1907, but without discussion.

587. *P. griseum* Bonorden, Abh. Geb. Myk., p. 92. 1864.

Colonies densely effused; hyphae gray, septate, flexuous, with apex slightly swollen and branched as in *P. glaucum*; conidia gray or greenish, larger than in *P. glaucum*.

Species reported from Westphalia as on rotten leaves, with the note

that the conidiophores were forked at the apex, with the branching in pairs. This species is given in Saccardo Sylloge 4: 78, in Lindau Rabh. Krypt. Lieferung 94, p. 164, 1904, in Westling Arkiv för Bot. 11: 147, 1911, by Wehmer in Lafar Bd. I. p. 375, 409, 419. Exsiccati under the name: J. F. Brenckle Fungi Dakotensis in Pathological Collections, U. S. Dept. Agr.

Gueguen in Bul. Soc. Mycol. France 14, p. 222, reports a *Penicillium* forming a gray colony on a solution of Tilia with conidia 3.5 by 2.1 $\mu$  hence corresponding with *P. griseum*, but becoming *P. glaucum* when transferred to other media. He decided therefore that these were not distinct species.

Biourge Monogr. La Cellule 33: 24, 1923, suggests this may have been Dierckx's *P. griseo-fulvum* but that it more probably was *P. digitatum* Sacc. on account of the large spores. No one therefore knows what Bonorden's species was.

588. *P. humicola* Oudemans. Arch. Néerlandaises der Sc. Exactes et Nat., 1902, p. 289, Tab. XXVI, fig. 1-5.

Colonies on gelatine azonate, orbicular, from cream to yellow green with creeping hyaline hyphae more or less floccose (spumoso), 1 to 4 $\mu$  in diameter; conidiophores hyaline, septate 110 to 120 by 1 to 1.5 $\mu$ , figured as coarsely sinuous, producing an apical verticil of 3 (?—C. T.) metulae 8 to 10 $\mu$  long noted as frequently curved or sigmoid; sterigmata also in threes, described as 5 $\mu$  long; conidia globose, hyaline, 2 $\mu$  in diameter.

Species found in cultures from humus from a forest in Holland by Koning. This species was afterward reported by Petri (Petri, L. Ueber die Wurzelfaule phylloxerierter Weinstocke Zeitschr. f. Pflanzenkr. 19: 25, 1909). The description by Oudemans specifies the penicillus as twice trichotomous. We are compelled to doubt the significance of or even the validity of the observations of metulae and sterigmata reported. We induced Koning a number of years later to go back to that forest and collect similar material, but were unable to recover this species in our cultures. It was certainly one of the biverticillate series and like *P. hirsutus* Bainier and Sartory in most of its characters.

Gilman and Abbott (no. 17, p. 292) report this species in Iowa. Their culture our No. 4894.9 is one of the biverticillate series perhaps near *P. pinophilum* but giving no close correspondence with Oudemans' description.

589. *P. incarnatum* Berkeley and Broome. Enumeration of the Fungi of Ceylon, in Jour. Linn. Soc. Bot. 14: 101, 1873; Sacc. Syll. 4: 84.

The data given follow: "No. 910. Pulvinulis minutis pallide carneis; floccis articulatis erectis hyalinis, apice digitatis; sporis limoniformibus (no. 241).

On leaves of some Monocotyledon. Spores .0003 (Saccardo gives 7 to  $8\mu$ ) long." The only possible clues to grouping are the "fleshy" color and the lemon-shaped conidia which suggest some member of the genus *Scopulariopsis*.

590. *P. macrosporum* B. & Br. Annals and Mag. of Natural History, Ser. 5, Vol. 9: 183. 1882.

The information given follows in full: "No. 1978. Aurantiacum, sporis globosis maximis. On a decaying Lactarius. J. D. C. Sowerby, whose drawing is in the collection of the British Museum."

No identification is possible from this publication.

591. *P. maculans* Sharples. Dept. Agr. Federated Malay States Bul. 19, p. 8. 1914.

Colonies upon sheet rubber becoming yellow in about 3 days, in some cultures, reddish brown; conidiophores upright figured as very short; sterigmata described as branched 6 to  $8.5\mu$  long, inadequately figured; conidia "globular" yellow to reddish brown, smooth  $2\mu$  in diameter; secondary spores egg-shaped 6 to  $8.3$  by  $5$  to  $4.5\mu$ , intercalary or terminal.

Plants found as the cause of spotting of plantation sheet rubber in the Federated Malay States. An effort to obtain cultures from the describer failed. Unless restudied and more completely described the species must remain unidentifiable.

- 591a. *P. maydis*. Lewin, L. Lehrbuch d. Toxicologie 2 auf. Berlin, 1897. Cited by Lafar. Handb. d. Technischen Myk. 2 auf. I, p. 613.

In Lafar, *P. maydis* is cited as designated by Lewin as a poison producing *Penicillium* in maize. No further data are given. The name *P. maydis* is not found in Lewin's book.

592. *P. megalosporum* Berkeley and Broome. *Annals and Mag. of Nat. Hst.*, Ser. 4, vol. 15: 34. 1875. See Sacc. *Sylloge* 4: 80.

The information as given is:

"No. 1457. "Niveum, breve; floccis fasciculatis; spores globosis elongatisque laevibus.

"In an old chicken coop. Menmuir, Rev. M. Anderson.

"Spores .0005-.001 inch (13 to 25 $\mu$  by Saccardo) in diameter, or equally variable when oblong."

No one has ventured to identify this species from the data given.

593. *P. microsporum* Rivolta. *Parass. Veget.*, p. 452. 1873.

Rivolta gives a five line note about a blue green mold without definite information which would furnish a clue to identity.

594. *P. minimum* Siebenmann. *Die Schimmelmycosen des menschlichen Ohres*, F. Siebenmann, Zweite vermehrte Ausgabe von: *Die Fadenpilze Aspergillus and Eurotium*. Wiesbaden, 1889, pp. 82-3.

Showing on membrane in ear of patient. Growing as blackish points in culture at 37°C., colorless when spread out under the microscope, but in mass as black as *Aspergillus niger*, mycelium 2 $\mu$  in diameter, smooth, colorless; conidiophore graceful, not ending in a vesicle but penicillium-like, without conidia (pinsellänge) about 20 $\mu$  in length; diameter of septa of conidiophore 6 $\mu$ ; conidia globose, smooth, 2.5 to 3 $\mu$ ; also clumps of entangled round, transparent, brownish cells, 20 $\mu$  in diam. which Siebenmann questioned as sclerotia or an impurity.

Isolated from acute otitis. No one has since claimed to recognize this species.

595. *P. monstrosum* Sopp. *Monogr.*, pp. 150-152, Taf. XVI, fig. 113; Taf. XXII, fig. 14. 1912.

Colonies on meat-peptone-sugar-gelatine, azonate with wrinkled and buckled mycelium, deep blue-green; reverse white with a bluish tint; no yellow color produced; rice alone became a brownish red color; odor none, or indistinct; conidiophores very coarse, and penicillus figured and described as showing either a single verticil of metulae or with branches at 1 or 2 nodes below the apical verticil, with metulae much swollen, obpyriform, and occasionally producing a secondary series similarly shaped; conidia elliptical to globose at times angular (!? CT), 4 by 3 $\mu$ ; perithecia not found.

Species obtained from cellar-earth; colonies showed an optimum temperature of 20°, minimum 5° and a maximum of 30°C.; they grew well on all the media and remained viable in culture for more than three years. No one has identified Sopp's species with certainty but from the description and figures we are compelled to believe that his cultures were contaminated or in some way pathological.

596. *P. montoyai* Castellani. Cited in Castellani and Chalmers, Manual of Tropical Medicine, p. 801, 1st edition, 1910, as of 1907 with *P. pictor* Neveu Lemaire, 1908, as synonymous.

Described in the "Manual" as conidia roundish or slightly oval, smooth, 3 to 4.5 $\mu$ , with cultures dark grayish color, and found by Montoya in cases of the grayish violet type of pinta.

597. *P. morsus-ranae* Corda. Icones V, p. 53, Tab. II, fig. 23. 1842.

Colonies described from rotting leaves of *Hydrocharis morsus-ranae* floating on the water, very delicate and evanescent, white, apparently velvety; conidiophores with stalk 1 to 3 celled, erect, colorless figured as rising from mycelium on the surface of the leaves, bearing at its apex a single verticil of 6 to 8 sterigmata, figured as diverging and producing long loosely tangled chains of conidia which break readily into separate cells; conida oval, hyaline 3 to 3.5 $\mu$  in long axis.

Species found at Prague, 1841, many times cited in the literature but not surely identified since.

598. *P. mycetomagenum* Mantelli and Negri. (Given in Gior. della R. Aco. di Medicina di Torino, ser. IV, 21: 165-166, 1915, as *P. mycetogenum* M. and N.)

Cited and the Latin diagnosis given as a footnote in Negri, G. Ricerche sulla biologia di un Penicillo patogeno, in Atti della reale Accademia della Scienze di Torino 56, disp. 4, 1920-1921, description pp. 67-68. 1921.

Parasite in melanotic granuloma of the human foot, Turin, Italy. Colonies when fruiting aeruginous green; floccose on bread, potato, carrot with sugar and glycerine, with white margin, finally murinus (mouse color). In gelatine fruiting between 20° to 25°C., growing slowly 3° to 37°, not liquefying.

Conidiophores crowded, mixed with sterile hyphae (not in coremia). Conidial apparatus several times branched, with terminal verticils of metulae 7.5 to 9.2 $\mu$  and sterigmata 3.5 $\mu$ , spherical depressed. Conidia

globose 2.2 to 3.7 $\mu$  smooth, hyaline, greenish aeruginous, never in long chains.

598a. *P. mycetomi* Neveu-Lemaire.

This name is given by Neveu-Lemaire (Precis de parasitologie humaine, p. 123, 4th edition, Paris, 1908) to a *Penicillium* which he claims both Brumpt and Bouffard noted independently. This *Penicillium* was recovered from a "mycetome du genou a grains rouges."

599. *P. ochroleucum* Artault. Recherches bactériologiques, mycologiques, zoologiques et médicales sur l'oeuf de poule, p. 213-4. 1893. Paris.

From air chamber of an egg. Clear yellow in egg and in culture, strong moldy odor, no especial characteristics, grew when placed near or on shell of egg in a moist place, penetrating readily pores of egg shell.

600. *P. pallidofulvum* Peck. New York State Museum Bul. 67, p. 30. 1903. Also in N. Y. State Museum Rep., 1902, p. 30.

Colonies described from rotting *Lactarius deceptivus*, as with mycelium densely tomentose creeping, pale tawny; conidiophores erect, septate, unbranched or with 1 to 3 short branches at the fruiting apex; conidia elliptical 3 to 4 $\mu$  in long axis, in chains.

Peck's material seems to have been lost. His description is entirely too meager to identify his species with new material. Until some one again collects specimens with substantially the same characters, the name must be placed among the unidentifiable species.

601. *Mucor penicillatus* Bulliard (Pierre). Herbar de la France, Vol. 3, pl. 504, fig. xi, 1791 (U. S. Dept. Agri., Library); Histoire des champignons de la France tome 1, partie 1, Paris, 1809. (The Farlow Library.)

Bulliard's figure is readily recognizable as some member of the genus *Penicillium* but offers no possibility of establishing a type species. The use of the generic term *Mucor* for groups having chains of conidia and septate mycelium was never retained.

602. *P. pezizoides* Biourge. La Cellule 36, p. 453-454. 1925.

Biourge mentions this name as assigned to his new no. 4 and in comment states that it is a striking fungus, developing in culture characteristic pedunculate cups each bearing a drop of liquid, which resemble a *Peziza*. Accordingly he assigns the provisional species name *pezizoides*.

His description of this form will be awaited with interest.

603. *P. pictor* Neveu-Lemaire. 1908.

Cited in *Precis de parasitologie humaine*, p. 89, 5th ed., Paris, 1921, as found by Montoya y Flores in grayish violet carate and stated by Sartory to have been provisionally used to designate a *Penicillium* described by Montoya y Flores in carate of the same type. Castellani reduced this species name to synonymy but Neveu-Lemaire restates its individuality as above in 1921.

604. *P. plicatum* Bon. Hanb., p. 75, fig. 81. 1851.

Bonorden described a gelatinous, plicate, mass or membrane about two lines thick upon the bung of wine cask filled with earth without regard to the purity of the mass.

Neither description nor figure furnishes any basis for recognizing this species although the figure indicates some species of *Penicillium*.

605. *P. poiraulti* Raciborski. In *Flora* 82: 119, 120. 1896. In article pp. 107 to 132.

Nomen nudum mentioned by the name on p. 119 and twice on p. 120, as growing in 1 per cent hydantoin, and as obtaining both nitrogen and carbon from 1 per cent hippuric acid.

606. *P. pruriosum* Salisbury.

The original citation and article have not been seen by the author.

Credited to Salisbury by Leon Marchand in *Botanique cryptogamique pharmacomedicale*, p. 194, fig. 45, a, b, c, d, 15°, 1883, and also by subsequent authors, who represent this material as having a greatly branched fruiting structure terminating in spores having a double contour (thick-walled?). Commentators state that Salisbury recovered this organism from vaginal mucus in severe pruritus and from the bladder of a man. This species is not identifiable.

607. *P. quadrifidum* Salisbury. *Zeitschr. Parasitenkunde* 4: 1-5, Taf. 1, fig. 1, a, b, c, d, e. 1875.

Salisbury's fungus was reported twice from the blood of erysipelas patients. As figured the conidiophores bore several sterile branches distinct from the terminal penicillus, consisting of a primary verticil of four branches, at first closely appressed, which after reaching a length four times their diameter formed septa and diverged to form metulae (?) each with a terminal verticil of four sterigmata and conidial chains. No measurements were given.



No one has ventured a guess as to the identity of this species from the incomplete description given.

608. *P. radians* Bonorden. Abh. a. d. Geb. Myk., 1864, p. 92.

The species was described as found on rotting leaves where it produced colonies blue green to gray fawn color, with fasciculate hyphae, short conidiophores with a single verticil of sterigmata and minute globose conidia. This was not sufficient information to permit the form to be again identified. It was cited by Saccardo Syll 4, p. 79, by Lindau in Rabh. Krypt. Lief. 44, p. 162 and by Westling, Arkiv. for Botanik 11: 146, 1911. No additional data have been furnished.

609. *P. rosato-fragens* Biourge. Monogr. La Cellule 33: fasc. 1, p. 225; Col. Pl. VI., Cart. 54; Pl. X, fig. 60. 1923.

Neither the discussion nor the figures warrant calling this more than a nomen nudum.

610. *P. rubro-punctatum* Dierckx. Soc. Scientifique Bruxelles 25: 85. 1901.

Colonies at first definitely blue then deep green, finally brown; reverse in wort cultures showing red spots, but showing light brown areas in Raulin's Solution; sterigmata 5 to 8 in number; conidia 2 to 3.5 $\mu$ .

The description was insufficient for identification; even Biourge with access to Dierckx's full notes and colored plates does not report this species.

610a. *P. rubrum* Sopp. Videnskapselskapets Skr. I. Mat.—Naturv. Kl. Kristiania 1911, no. 2. pp. 19–20, Taf. 5, fig. 28–29. 1912. Also fig. 131, Taf. XVIII in same Skr. No. 11. 1912.

Colonies at first gray-green, later gray, with long slender conidiophores bearing very numerous sterigmata producing abundant oval hyaline very small conidia which appear quickly in cultures and give the colonies a reddish color. Perithecia red, about 0.5 mm. in diameter, truffle-like, in morphology and habit like the same structures in *P. glaucum* as described by Brefeld (hence no description was given!). Cultures remained viable only one year. The species grew best upon flesh, less well upon bread and upon wood, poorly upon gelatine media, not at all upon agar; it did not grow above 25°C.; it produced very slight odor. In old cultures only "ascusfrüchte" were found; no conidia.

Sopp regarded the conidial stage as connected with a parasitic phase of his organism upon insect larvae and admitted that his culture appeared to be parasitized (impure!—C. T.). Detailed description was reserved for his monograph but not given there under this name although his *P. niveorubrum* has suggestive resemblances in description. No one has since identified it.

611. *P. silvaticum* Oud. Fl. Myc. obt. sur gelatine, etc. Archives Neerlandaises des sc. Exactes et Nat. 1902, p. 289, tab. XXVII, fig. 1-4.

Colonies avellaneous, not zonate, velvety in appearance, with conidiphores up to  $210\mu$  long by  $2$  to  $3.3\mu$ , bearing a single verticil of sterigmata  $12$  to  $22\mu$  long with long chains of conidia globose, smooth  $2.3\mu$  in diameter.

Isolated by Koning from leaf mold in Holland.

Culture; by Koning and Oudemans. Not definitely identified by us. From its avellaneous color, and the excessive length of the sterigmata we have always suspected that the type material might have been *Aspergillus terreus* Thom, in which the double series of sterigmata is sometimes detected with difficulty.

612. *P. socium* listed and credited to Plowright in Saccardo's Sylloge 2: 468, 1883, as the conidial stage of *Hypomyces aureonitens* Tulasne, redescribed by Plowright (Grevillea 11: 49, tab. 156, figs. b, d, f, 1882). This name is also listed by Grove (Jour. Bot. 23: 165, 1885) as a synonym for *Gliocladium penicilloides* Corda.

Plowright figures and gives measurements for a conidial form of *Hypomyces aureo-nitens* Tulasne without suggesting the allocation of this to Penicillium: Penicillus as figured and described, triverticillate in figures, with conidia described as minute, hyaline, oval (elliptical as figured),  $3$  to  $4\mu$  by  $2\mu$ , in short chains.

Plowright's material came from a rotting *Stereum hirsutum* in Wales and involved no culture to insure a relationship between his Hypomyces and the Penicillium-like conidial masses. Without further data Saccardo's name *P. socium* may be dropped whether the organism belongs with Gliocladium as suggested by Grove or not.

613. *P. subtile* Berkeley, no. 241. Annals and Magazine of Natural History, Ser. 1, 6: 437, Tab. XIV, fig. 25. 1841. See Cooke, Handbook, p. 603. Also Sacc. Sylloge 4: 80. 1887.

Colonies "clothing the inside of an old willow," as a very fine delicate, mealy "bloom" of creeping hyphae and simple or branched erect conidiophores (sometimes ternate), producing a few chains of rather large broadly elliptical conidia apiculate at both ends.

Berkeley figured the conidiophores as septate and thick walled; the conidia as heavy walled, and rounded at both ends, and as arising singly or in groups at the apex of the conidiophore or at septa below the apex. The species can not be identified as a *Penicillium* from either description or figure, hence may be excluded.

Collected at Tansor, Norths, England, in spring.

614. *P. subtile* var. *ramosius* Grove, W. B. New or Noteworthy Fungi: Part II, Journal of Botany 23: 165. 1885.

Colonies on rotten wood entirely white, delicate; conidiophores erect, commonly ternate, with a few branches below the apex; conidia colorless, broadly elliptical 16 to 20 by 10, apiculate at both ends, in chains of 4 to 8.

Found at Hampton-in-Arden, England, on rotten wood.

Grove's description is in Latin, but without figures. His notes on other forms indicate close relations with Plowright. Biourge cites this organism as *P. ramosius* in his "List onomastique." Reference to *P. ramosius* Greville is probably a misinterpretation of the abbreviation Grov.

615. *Coremium syphiliticum* Hallier, in Flora 51: 295, 301, fig. 16, 1868, is described as a form assumed together with *P. syphiliticum* by *Coniothecium syphiliticum*. This name must be discarded as noted for *P. gonorrhoeicum* Hallier.

Associated by Hallier with syphilis.

- 615a. *P. syphiliticum* Hallier, in Flora 51: 295, 301, fig. 12, 1868, is described as a penicillate form (Prap. nr. 369) assumed by *Coniothecium syphiliticum*. As noted under *P. gonorrhoeicum* of Hallier this name must also be discarded.

Associated by Hallier with syphilis.

616. *P. tenellum* Cooke. Grevillea 7: 15, September, 1875.

Cooke's material consisted of rotting leaves of *Symplocos* from Bengal, India, bearing patches of pale fumose mold 20 to 30 mm. in diameter with conidiophores scarcely  $100\mu$  long, simple or branched, septate, and diaphanous. The conidia were  $3\mu$  in diameter, globose, and hyaline. Identification is impossible.

617. *P. toruloides* Preuss. Linnaea 25, p. 729, 1852.

Colonies coarse, flesh colored, erect hyphae "carneo-rubris," few branched with branches more or less diverging, chains of conidia few; conidia globose, on badly dried beans.

Possibly a *Scopulariopsis*. Not identifiable.

618. *P. verticilliferum* Spegazzini. Physis 7, no. 23, p. 18, 1923.

Colonies found upon gum in the leaf axils of *Nothofagus antarctica*, whitish cinereous, with vegetative hyphae creeping and bearing scattered or loosely clustered conidiophores, 30 to 50 by 6 to  $7\mu$ , irregularly branched, very short with apex abruptly bearing upright branches 10 to 15 by 5 to  $6\mu$ , these in turn bearing a corona (whorl) of five to eight elements, 250 to 300 by 4 to  $5\mu$ , having five to seven septa each, these elements bearing one to three symmetrical branches, which in turn are one or twice bifurcate at the tip with various, septate sections superimposed and becoming insensibly transformed into conidia, catenulate, at first elliptical then globose, 4 to 6 by 4 to  $5\mu$ , smooth, hyaline.

Spegazzini evidently mounted little tufts of mold, found upon the dry masses of gum in his specimens, and described as elements in a penicillate fruiting organism the branching system at the base of the tuft under observation. While no identification can be offered it is entirely probable that under modern systems of culture his species would have proved to be some ordinary green *Penicillium* with rather large conidia.

619. *Penicillium virellum* Peyronel (B) in I germi atmosferici dei funghi con micelio, page 22, Padova. 1913.

Colonies discoid, zonate, at first white, presently green (viridis); sterile hyphae septate, hyaline, complexly branching, 3 to  $4\mu$  in diameter; conidiophores erect, rather short, 40 to 60 by 4 to  $6\mu$ , septate, with apex pale green, penicillately branched; branches paired or ternate 14 to 20 by 4 to  $6\mu$ , with apex 3 to  $5\mu$ ; sterigmata verticillate, 12 to 15 by 3 to  $5\mu$ , pale green, producing conidia in long obconical masses; conidia globose or broadly elliptical 3.5 to 4 by 3 to  $3.5\mu$  green smooth.

Habitat: Air in the open fields; cultivated in onion infusion at Padua, Italy. This "species with no aerial mycelium and short conidiophores" might belong with such species as *P. atramentosum* Thom no. 130a, *P. oxalicum* Currie and Thom no. 120.

## CHAPTER XXVI

### SPECIES OF OTHER GENERA DESCRIBED AS *PENICILLIA*

Changing concepts of generic limitation, during the one hundred and twenty years since Link published his "Observationes" have put many species in and again out of the genus. Many of the earlier workers finding conidia, forming a cluster of chains at or about the tip of a conidiophore described their organisms as *Penicillia* without recognizing two markedly divergent types of chain. In the *Penicillium* type, each successive conidium is produced directly upon the tip of the sterigma, pushing the next older one outward so that the oldest one is at the end of the chain. In the other, the oldest cell is at the base of the chain, buds to produce the second, occasionally buds twice to produce two chains, and each succeeding cell is capable of similar budding so that the newest cells are at the tips of the chains or of branching systems of chains.

Even within the group of organisms in which the sterigma is a specialized conidium-producing organ there is so great divergence in other structures that generic lines are still uncertain. Certain species have been separated because obviously belonging elsewhere; probably other species should be separated.

The species believed definitely to belong in genera not otherwise considered in this book are given here in alphabetical order to the names used in designating them as *Penicillia*.

630. *P. abnorme* B. and Br. No. 1914 of British fungi. M. J. Berkeley and C. E. Broome, Ann. and Mag. Nat. Hist. 7, 5th series, p. 130, pl. iii, fig. 4. 1881.

The entire description reads: "Candidum, floccis tenuibus in corpus turbiniforme desinentibus, sporis minutissimis." The figures show distinct chains of spores extending out of an urn-shaped, stalked receptacle.

Habitat: Leaves of *Trientalis europaea*.

Not a member of the genus *Penicillium* in the sense of this paper.

631. *P. albo-marginatum* Biourge. In Monogr. La Cellule 33: fasc. 1, Col. Pl. XIII, Cart. 24; Pl. XXII, fig. 129. 1923.

Synonym: *Aspergillus albo-marginatus* Biourge.

No description was given for this species as a *Penicillium*; the figures were included in the monograph and labeled *Penicillium*. Cultures re-

ceived in August, 1928, were labeled *Aspergillus albo-marginatus* Biourge no. 609 (no. 4733.134) and *Microaspergillus albo-marginatus* Biourge (no. 4733.134a). These cultures certainly belong in the *A. conicus-gracilis* series of species.

632. *Amblosporium* Oudemans and Koning.

In Biourge Monogr. La Cellule 33: fasc. 1, p. 323, this is noted as a *Microaspergillus* and excluded from *Penicillium*.

633. *P. armeniacum* Berkeley. Introduction to Cyptoganic Botany 1857; p. 298, fig. 68c.

This species as figured was not a *Penicillium* but a form with branching chains of conidia but giving no basis for closer identification although the species has been reported by others as *Monilia sitophila*.

634. *P. atrobrunneum* Cooke. Grev. 6: 139. 1877-8, Rav. Fung. Amer. no. 59.

The description given reads: "Elongato-effusum, atro-brunneum; hyphis erectis, sparse ramosis, septatis, apice bi-vel tri-furcatis; sporis elongato-ellipticis (0.008-0.01-0.004 mm.)."

On leaves of Musa. Gainesville, Fla.

Synonym: *Haplographium atrobrunneum*. Sacc., Syll. 4: 305, 1886.

Synonym: *Schizocephalum atrobrunneum* (Cooke) Pound and Clements in Bul. Geol. Nat. Hist. Survey Minnesota 9 (Bot. Ser. II): 666, 1894-1898.

635. *P. (Oospora) auridorsum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 228; Col. Pl. XII, Cart. 303; Pl. XXI, fig. 121. 1923.

Colonies on wort gelatine at first chalk white to pale yellow; reverse beautiful golden brown; gelatine quickly liquefied; odor ammoniacal, or arsenical; conidiophore simple or bifurcate, 2 to 3 $\mu$  in diameter; whole conidial apparatus reduced to a short stalk bearing a conidial chain or once forked hence producing two chains; conidia 5 to 6 by 3.5 to 4.5 $\mu$ .

Biourge's no. 303 (our no. 4733.13) grew in Czapek's solution agar as small yellow to orange colonies producing a bristly surface, hyphae in mass up to 300 $\mu$  deep, reverse bright yellow to orange; agar little colored. The organism in our cultures could not justly be included in *Penicillium* or in *Scopulariopsis*.

636. *P. bouffardi* Brumpt.

Evidently a typographical error for *Aspergillus bouffardi* Brumpt, copied and handed down over a period of time.

637. *P. brevipes* Corda. Icones IV, p. 31, Taf. VII, fig. 93. 1840.

This was described as a gray mold on the peeled wood of *Sambucus nigra*. The figure and description suggest an *Aspergillus* such as *A. nidularis* rather than a *Penicillium*. Both figure and description lack the data necessary for identification.

639. *P. canum* Preuss. Linnaea 24: 135. 1851.

The description reads: "Caespitibus minutis cano-fuscis, late expansis, pulverulentis; hyphopodio ramoso conjuncto, rarius septatis, ramis verticillatis; floccis sporarum longis solitariis; sporis (magnis) subovatis, episporio hyalino ferrugineolo, utrinque hilo instructo; nucleo granulato."

Habitat in foliis Allii Moly putridis post pluvias in hortis. Hoyer-swerda.

No identification is possible from Preuss' description, although Hanzawa, (Hanzawa, J. Myc. Centralb. 5 (1914), 1, pp. 4-13) uses it for a *Penicillium* upon onions, with colonies small, greenish blue; conidiophores little branched 290 by 3.2 $\mu$ , with verticillate sterigmata up to 16 by 3 $\mu$  and conidia finely punctate, 3.2 by 2.4 $\mu$ . There does not appear to be satisfactory reason for this identification, and we are inclined to believe Preuss did not have a *Penicillium* in the first place.

640. *P. caulatum* Sopp. Monogr. p. 103, Taf. II, figs. 7-12. 1912.

Mycelium consisting of brown, coarse, firm hyphae forming loose networks or coarse felts, showing to the naked eye darker, denser clumps recognized as sclerotia under the handlens; from this mycelium the conidiophores arise like brown bristles; conidiophores arising from bulb-like bases or from sclerotia, septate, brown walled, becoming paler above or seeming growing out from a sheath, large, coarse, warty, with thick walls up to 800 $\mu$  by 20 $\mu$ , somewhat narrowed above up to six times (verticillately) branched with the lowest branches coarser and darker (with brown walls) than the upper branches, and from the figures progressively reduced in diameter; sterigmata very slender and sharp pointed (needle-like); sclerotia in large clumps surrounded by warty, brown hyphae from which conidiophores arise; perithecia small, green or greenish-brown with a thin parenchyma-like wall and asci with globose



medium-sized ascospores were reported but later noted as not definitely shown to belong in this species; conidia small, elongated, uniform or comma form (bacterium-like), about 0.5 by  $1\mu$  hyaline, produced in great numbers, germinating to produce very delicate hyphae which only gradually enlarge.

Fungus discovered by Sopp upon a fallen twig of pine in the soil, and visible as forming small brown bristles upon the wood; colonies grew poorly upon gelatine media, but developed slowly upon media prepared with an extract of pine and older twigs; optimum temperature  $15^{\circ}$  to  $20^{\circ}\text{C.}$ ; odor slight somewhat mosslike.

Sopp's description fails to give essential measurements of perithecia and ascospores or to show the connection of the perithecia reported with the conidial fungus described.

Three cultures, nos. 81918.1, 5118.4, and 102518.1, obtained from wood by C. J. Humphrey of the Office of Forest Pathology resemble *P. caulatum* in many characters. They are certainly not to be included in *Penicillium*. Miss Caroline Rumbold furnished a culture to the American Type Culture Collection no. 1770 labeled *Graphium penicilloides* which appears to satisfy Sopp's description.

641. *P. chartarum* Cooke. Popular Science Review 10: 30-31, Pl. LXVIII, fig. 4. 1871.

Commonly olivaceous to dark olive, sometimes dirty reddish, with erect septate hyphae, branching toward the upper portions in a fasciculate manner and these branches bearing chains of spores forming a tassellike head; spores oblong.

Found on varnished, marbled wall paper, the varnish being pushed off in translucent flakes by the mold growth.

Synonym: *Haplographium chartarum* (Cooke) Sacc., Syll. 4: 305. 1886.

642. *P. cicadinum* V. Höhnelt. Sitzungsber. der kaiserl. Akad. Wissensch. Wien Mathem.-naturw. Klasse 118, Abt. 1, (pp. 131-133 in reprint) 1909.

On large singing cicadas white, then clear blue green and finally olive green from conidial mass; fruiting hyphae branched, bushy, colorless, smooth, 2 to  $3\mu$  in diam. at upper end of sterigma; chains of conidia more than  $100\mu$  in length, parallel, massed; conidia rod-shaped, rounded at ends, 5 to 6 by 1.5 to  $2\mu$ , rarely  $7\mu$  (germinating?).

V. Höhnelt states that this form may be *Oospora* rather than *Penicil-*

lium because the branching of the fruiting hyphae is not typically penicillate.

Pathogenic to cicadas.

Slide in V. Höhnelt collection, Farlow Herbarium labeled H.410, Buitenzorg, 1908, shows conidia yellowish green, of the *Metarrhizium* group.

643. *Syncephalastrum cinereum* Gueguen.

In Biourge Monogr. La Cellule 33: fasc. 1, p. 323, Pl. XXIII, fig. 135; 1923; this is noted as a *Microaspergillus*, with short stalks arising from trailing branches, and elliptical slightly roughened spores (hence suggestive of Bainier's *A. gracilis*.—C. T.).

644. *P. chlorinum* Fresenius. Beitr. z. Myk., p. 22, Taf. III. 1850.

"Die Sporen sind hellgrün, rund, 1/186 mm. gross. Tafel III. Figur 20 sind Theile des Sporenstandes und Sporenketten (trocken) abgebildet, Figur 21 einzelne Sporen." The next line reads: "Eine gleichfalls verwandte Art:" followed by the name and descr. of *P. cladosporioides* Fresen.

The figure suggests a *Cladosporium* or *Hormodendrum*.

644a. *P. chlorocephalum* Fresenius.

In Biourge's List onomastique appears to be Fresenius' *Periconia* in Fresenius Beitr., p. 20, 1850. Figure and description exclude it from *Penicillium*.

645. *P. cladosporioides* Fresenius. Beiträge zur Mykologie, Taf. III, pp. 22–23, figs. 23–28. 1850–3.

Fresenius gives a series of figures which clearly place this form with *Cladosporium*.

6. *Oospora crustacea* Bulliard. In Biourge, Monogr. La Cellule 33: fasc. 1, p. 229–230; Col. Pl. XII, Cart. 299; Pl. XXI, fig. 125. 1923.

? *Sporendonema casei* Desmazieres, also Syn. *Hyalobysus moniliforme* Zukal.

Biourge no. 299 (our no. 4733.46) is recorded as the "rouge du fromage." The figures are admitted to have been "deformations" due to age. The organism is recorded as an "arsenical." The carton Col. Pl. XII,

299, (given as no. 206 on p. 230) is regarded as enough to identify the species. Our cultures upon Czapek and wort agar show colonies at first submerged, then producing a loosely floccose mass up to 1 mm. deep in age with pale yellow fruiting zone near the margin; spore production taking the form of chains of oidia; other chains and areas of cells irregularly enlarged are common.

There is no suggestion of relationship to *Penicillium* in this culture.

646a. *P. curtipes* Berkeley. In Ann. and Mag. Nat. Hist., Ser. 2, Vol. 2, pp. 380-383, Pl. XI, fig. 1848.

Berkeley's figure and description were based upon exsiccati of Karl Thomas. The chains of conidia were distinctly described and figured as branching hence the species was not a *Penicillium*.

647. *P. echinatum* Rivolta. Parass. veget. p. 451, Tav. VI, fig. 150-151; Synonym: 1873. *Haplographium echinatum* (Riv.) Sacc.

Examination of Rivolta's figures and description shows that Saccardo correctly took this organism out of the genus *Penicillium*.

647a. *Citromyces exiguus* Bainier and Sartory in Biourge's List Onomastique is apparently a mistake for *A. gracilis* var. *exiguus* of their 1912 paper.

648. *P. finitimum* Preuss. In Fungi Hoyerswerda no. 118, Linnæa 24: 134. 1851.

Preuss' statement that the base and lower part of the conidiophore were intensely dark walled (atro-fuscus) excludes his organism from *Penicillium*. Saccardo (Syll. IV, 307) placed it in *Haplographium*. Von Höhnelt (in Sitzungsber. Kais. Akad. Wiss. Wien, II Klasse 115: 649-695, 1906) reported that Preuss' three species *P. fuscipes*, *P. finitimum* and *P. flexuosum* all found upon pine needles are identical and should take the name *H. finitimum* (Preuss) Sacc.

649. *P. firmum* Preuss. In Fungi Hoyerswerda, Linnæa 24: 136. 1851.

The description lacks essentials for identification of any species, while the specification of fuscous stalks excludes the material from *Penicillium* definitely. No subsequent author has identified or discussed this species from culture or exsiccati.

650. *P. flavo-virens* Cke. and Mass. Grev. 20: 106. 1891.

Dense, effuse, floccose, flavo-virens with creeping, interwoven hyphae, bearing suberect fruiting hyphae, bifurcate at apex and strict. Conidia in chains, elliptical, minute, hyaline, 3 to 4 by  $1\mu$ .

On fruits of *Terminalia belerica*, Ceylon.

Thwaites, 374.

Not recognizable even as a *Penicillium* in the sense of this paper.

651. *P. flexuosum* Preuss. Linnaea 24 (1851), p. 135, F. Hoyer. no. 119.

Preuss specifies "hyphopodio stratosata atro-fusca" and "stipite erecto infra atrofusca supra pallido." While not offering a basis for identification this excludes *P. flexuosum* from *Penicillium* as discussed here. Saccardo places it in *Haplographium* (Syll. IV. 307).

651a. *P. fulvum* Rabenhorst. Krypt. Fl. 1 Aufl. 1, 92. 1844, was given as a synonym of *Rhodocephalus aureus* Corda Icones III, p. 12, fig. 33. 1839.

Corda's figure might have been an *Aspergillus* or a monoverticillate *Penicillium*. The probability is that it was some *Aspergillus* like *A. terreus* Thom.

652. *P. fuscipes* Preuss, Linnaea 24 (1851), p. 136, F. Hoyer. no. 123. Synonym: *Haplographium fuscipes* in Sacc. Syll. IV: 307, and Centralb. f. Bakt. 2 Abt. 20: 178.

The specification hyphopodium fuscous, with lower part of stipe also fuscous, appears to eliminate this species from *Penicillium*. Von Höhnelt (Stizungsber. Kais. Akad. Wiss. Wien II Kl. 115: p. 649-695, 1906) regarded this species as identical with *P. finitimum* q.v.

652a. *Penicillium gonorrhoeicum* Hallier. In Flora 51: 294, 300, fig. 9, 1868, is described as a penicillate form (Prap. nr. 342) assumed by *Cladosporium gonorrhoeicum*.

Hallier in the course of a five years' study of pathological material prepared over a thousand mounts. His drawings were evidently made from these mounts. This species name is to be discarded for lack of any accompanying description. Associated by Hallier with gonorrhea.

653. *Trichurus gorgonifer* Bainier. Bul. Soc. Mycol. France 23: 229-233; Pl. XXV, fig. 1-6. 1907.

*Trichurus* (Clements and Shear, Bot. Surv. of Nebraska, Lincoln, 1896, p. 7) is a hyphomycete genus, belonging to the Rhaeostilbeae and having penicillus-like conidial apparatus produced at first sparsely over an extended mycelium, later upon vertical cylindrical stilbum or Isaria-like coremia, and separated from *Stysanus* by the production of filaments among the fertile elements of the penicilli.

*T. gorgonifer* as described: colonies at first white tufted masses, becoming progressively gray to almost black; coremia solitary or in tufts, with stalks up to  $2.5\mu$  in height,  $35-85\mu$  in diameter, rigid, simple or double, black at base, slightly attenuate toward the apex and enlarging to form a head, cylindrical clavate, at times narrowed toward the base, covered with radiating, curved, or S-shaped hairs, sharp pointed, dark brown, septate,  $60$  to  $120\mu$  or even  $200\mu$  by  $2$  to  $2.5\mu$ ; conidial apparatus at first sterigmata and groups of sterigmata scattered along simple hyphae, or on the tips of short fertile branches, then pericillate masses on variously branched simple hyphae, and later upon similar branches from fertile heads of coremia and producing hairs as sterile proliferations of part of the sterigmata or metula-like branches, thus giving the characteristic appearance of the species; conidia oblong elliptical from  $5.6$  by  $2.8$  to  $8$  to  $9$  by  $3\mu$ , green, smooth. (Bainier's two statements do not agree since conidia are  $8$  to  $9$  by  $3$  on p. 229 and  $5.8$  by  $2.8$  on p. 233.)

Type found upon cow dung grows readily on paper, straw, licorice root, better on solid than on liquid media.

654. *P. griseo-atrum* Biourge. Monogr. La Cellule 33: fasc. 1, p. 301; Col. Pl. XI, Cart. 411; Pl. XIX, fig. 113. 1913.

Colonies on wort gelatine; at first conidial margin bluish green, within gray green, then gray, and finally dark gray to fuliginous; coremia none; reverse pale yellow to reddish orange; odor none; conidiophores  $50$  to  $130$  by  $0.5$  to  $5\mu$ , gradually enlarging from the base upward, with all walls smooth, figured as arising at almost a sharp point; sterigmata  $8$  to  $12.5$  by  $3.2$  to  $5\mu$ , in verticils for  $5$  to  $12$ ; conidia elliptical  $2.2$  to  $3.8$  by  $1.8$  to  $3$ .

Biourge type no. 411 was not received: Figures and description of stalks suggest an *Aspergillus*, the colors and medallions suggest *A. conicus* of Blochwitz. At least two of Biourge's cultures as received contain members of this series hence it is certain that the species was

present in his laboratory. A culture numbered 411 was received in 1927 but was not this species as described.

655. *P. Guegueni* Biourge. In Biourge Monogr. La Cellule 33: Fasc. 1, Species list, p. 103, and in Col. Pl. XIII, Cart. 23; Pl. XXII, fig. 128, 1923.

Synonym: *Aspergillus Guegueni* Biourge.

No description was given in Biourge's monograph. The figures were printed over the label *P. guegueni*. Cultures received from Biourge in August 1928 labeled *Aspergillus guegueni* (4733.139) and *A. (Microaspergillus) guegueni* (4733.139a) agree with the figures in placing this organism in a series with *A. gracilis* Bainier.

656. *P. (Microaspergillus) Hickeyi* Biourge No. 15. In Monogr. La Cellule 33: fasc. 1, p. 103, 323, 328, and in Col. Pl. XIII, Cart. 15, Pl. XXII, fig. 127. 1923.

Synonym for *Aspergillus Hickeyi* Biourge.

No description was given in Biourge's monograph but the figures cited are labeled as *Penicillium*. Two cultures received from him in August 1928, were labeled *Aspergillus Hickeyi* (no. 4733.141) and *A. (Microaspergillus) Hickeyi* (no. 4733.141a). Both are *Aspergilli* and belong in the section with *Aspergillus conicus* Blochwitz and *A. gracilis* Bainier.

657. *P. hypo-janthinum* Biourge, No. 25. In Monogr. La Cellule 33: fasc. 1, p. 321-322; Col. Pl. XIII, Cart. 25; Pl. XXII, fig. 130. 1923.

Synonym: *Aspergillus hypojanthinus* Biourge.

Colonies on wort gelatine restricted in growth, almost velvety, wrinkled (contorted) yellow to ochraceous, figured as having a surface growth of interlacing ropes of hyphae with conidia scantily produced hence few green areas; coremia none; reverse ochraceous yellow, to purplish; gelatine rapidly liquefied; odor none; conidiophores very short branches from trailing or ascending hyphae and ropes of hyphae, 15 to 40 by 1.5 to 2.5 $\mu$ , sometimes enlarging at apex to 4 $\mu$ , with all walls smooth; sterigmata 6 to 10 by 2.5 to 3 $\mu$ , in groups of 3 to 6, figured as diverging at the apex and bearing divergent chains of conidia; conidia oblong 2.5 to 3 by 2 to 2.4 $\mu$ .

In August, 1928, we received two cultures from Biourge labeled *Aspergillus hypojanthinus* (no. 4733.142a) and *A. (Microaspergillus)*

*hypoanthinus* (no. 4733.142). These appear to be the *P. hypoanthinum* described above and are correctly placed in *Aspergillus* near *A. gracilis* Bainier.

In September, 1927, we received a culture from him numbered 25 (our no. 4733.72) which agrees essentially in removing this species to the *Aspergilli*.

658. *P. hypomyces* Saccardo Sylloge 4: 80.

Synonym: *Hypomyces aurea-nitentis*. Plowright Greville 11: 49; tab. 156, fig. e, d.

Saccardo gives: Effused, white; conidiophores erect septate hyaline, with conidial apparatus dichotomous or trichotomous. Conidia ellipsoid, hyaline 3 to 4 by 2 $\mu$ .

Species found upon *Stereum hirsutum* in Great Britain, resembling *P. candidum* but differing in the elliptical conidia and more regular branching.

659. *Scopulariopsis leproides* Leger and Nogue. Bull. Soc. Path. Exot., Paris 15: 654-661, figs. 1-3. 1922.

Colonies on Sabouraud medium maturing in five to six days at 20° to 26°C., but developing more rapidly at 37°C. At first dirty white, then buckling and becoming green to bottle green to slatey gray, not liquefying gelatine, blackish gray on potato, not coagulating litmus milk, fermenting lactose rapidly, but not forming acid in maltose and glucose. Mycelial filaments 2.5 to 4 $\mu$  in diameter, septate, branching; conidiophores commonly 20 to 60 $\mu$  long, bearing at the slender apex an oval mother conidium which cuts off several daughter conidia; other conidia borne at various levels, alternate or verticillate, and in false tufts, but not resembling a head of *Aspergillus*; conidia 4 to 6 $\mu$  in long axis, thick-walled and described bearing two elevations near one end of the conidium.

This description would exclude the species from *Scopulariopsis* as we have encountered the genus, both by its green color in some cultures and by the method of conidium formation described. Unfortunately, we have not seen the original.

Leger and Nogue claim to have recovered this fungus from a dermatomycosis of the forearms and hands of two mussulmen who always struck the ground on salaaming, the suggestion being an infection through soil contamination.

659. *P. leucocephalum* Rabenhorst. D. C. Fl. n. 857; *Rhodocephalus candidus* Corda, Icones fungorum I, p. 2, fig. 282.

Corda's figure shows a mold with conidia in branching chains which excludes it from *Penicillium*.

660. *P. lobulatum* Bon. Abh. Geb. Myk., p. 92, 1864.

Synonyms: *Sporocybe lobulata* Berkeley; *Rhodocephalus* Corda.

Distributed as *P. lobulatum* in Rabenhorst's Fungi Europaea no. 171.

Although Bonorden regarded this as a true *Penicillium*, his recognition of black hyphae and oval black conidia excludes it from our conception of the genus.

661. *P. nigrovirens* Fresenius. Beitr. z. Myk., p. 22, Taf. III, fig. 22.

Fresenius recognized a close relationship of this material with that described by him as *P. chlorinum*, *P. viride*, and *P. cladosporioides*, all of which belong in *Cladosporium* or *Hormodendrum*. It was found also on rotten fruits, but was dark olive green instead of the clear green of *P. chlorinum*.

The figures and descriptions clearly remove this species from *Penicillium*.

662. *Penicillium oidiforme* Orlova. Journ. Soc. Bot. Russia 10, nos. 3-4, pp. 375-394, 8 figs. 1925 (In Russian with French résumé).  
Published in 1926.

Single spore isolations when cultured on different media containing combinations of beef extract, peptone, glucose or lactose with gelatine or agar and liquid media containing nitrogen, carbon, phosphate, magnesium sulphate with in addition citric acid, sulphuric acid, hydrofluoric acid or lactose developed colorless hyphae, 4 to 5  $\mu$  in diameter, separating easily into oidia, with individual cells 5 to 6 nucleate; stalks, 5.8 to 7  $\mu$  in diameter, varying in length with wall verrucose, pimpled (in Russian and as pictured); metulae 4 to 4.5  $\mu$  by 10 to 13  $\mu$ , verrucose as walls of conidiophore; sterigmata, 2.9 to 3 by 8 to 10  $\mu$ , smooth, not always separated from the metulae by septa, sometimes separated by an internal annular ring; conidia globose, 4.5 to 4.8 (5)  $\mu$  in diameter, one-nucleate, grayish green in mass, sometimes formed before the metulae.

Optimum temperature 14° to 16°C.

Oidia developing in cultures having a pH of 2 to 8.3, mycelium at extremes of pH as 1.5 to 9, yeastlike forms in cultures having (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>



and  $\text{NH}_4\text{NO}_3$  as a source of nitrogen and upright "gemmes" in the presence of lactose. The most beautiful cultures developed with sucrose in the medium.

Perithecia unknown. As cultured and described this form is not recognizable as a *Penicillium* in the sense of this monograph.

663. *P. olivaceum* Corda. Icones 3: p. 12, Taf. II, fig. 35. 1839.

This was some species of *Cladosporium* or related form with conidiophores colored and branching spore chains, described from colonies found upon birch wood, without culture or sufficient data for subsequent identification.

664. *P. olivaceum* var. *discoideum* El. and Em. Marchal. Bul. Soc. Roy. Bot. Belg. 54: p. 129. 1921.

Latin diagnosis translated: hyphae separate, more frequently remarkably united into a disc, 4.5 mm. in diameter and 1 to 2  $\mu\text{m}$ . in height; conidia ovoid or elliptical, hyaline, in mass olivaceous, smooth 3.6 to 6.8 by 2.3 to 3  $\mu$ , in long chains presently deciduous. Habitat on fruits of *Prunus*.

From the description this form was probably *Metarrhizium*.

665. *P. orbicula* Corda. Icones 3: p. 12, Taf. II, fig. 34. 1839.

Synonym: *Briarea orbicula* (Corda) Bon. See Saccardo Syll. IV: 85; XI; 668.

Corda's figure is idealized beyond recognition. In his descriptive notes concerning colonies on rotting paper, one familiar with the *Cladosporium* group in such situations finds evidence that some member of that series furnished the material for both figures and description. Forking chains of conidia are both figured and described.

666. *P. ovoideum* Preuss in Fungi Hoyerswerda 272, Linnaea 26: 708. 1853.

Colonies diffuse indeterminate white, with mycelium stratose forming a submerged or close lying mat of hyphae; conidiophores erect, colorless, with verticillate branching at the apex and the the branches bearing groups of branchlets and spore chains; conidia ovoid white; found upon moist isinglass. Hoyerswerda.

No identification is possible.

667. *P. pertardum* Biourge. In Monogr. La Cellule 33: fasc. 1, Col. Pl. XIII; Cart. 27; Pl. XXII; fig. 132. 1923.

Synonym: *Aspergillus* (*Microaspergillus*) *pertardus* Biourge.

Biourge figured this species in the monograph but did not describe it as a *Penicillium*. In July, 1928, he sent us two cultures, one marked *Aspergillus pertardus* Biourge Emend. no. 27 (our no. 4733.144a), the other *A. (Microaspergillus) pertardus* (no. 4733.144). The organism is certainly an *Aspergillus* and may be placed next to *A. penicilloides* Spegazzini from which it differs by its large conidia.

668. *Penicillium polyactis* Secretan. Mycographie Suisse 111: 537. 1833.

This name was regarded by Secretan as a synonym of Link's *Polyactis vulgaris* and Persoon's *Monilia polyactis*, and the description attached would exclude *Penicillium* in the sense of this book.

- 668a. *Monilia racemosa* Pers. Syn. Meth. Fung., p. 692, and based upon Micheli Pl. 91, fig. 4.

Cited as a *Penicillium* by Hoffman from Westendorp's Herb. Crypt. No. 196. Persoon's *Monilia racemosa* is not determinable as a *Penicillium*.

669. *P. radiatum* Lindner. Mikroc. Betriebskontr., 1901, p. 314; also *ibid.*, 1909, 5 aufl., p. 383-384, fig. 164.

Found and figured from berries of *Vaccinium*, then cultivated. Lindner's preparation of the original material preserved in his laboratory is certainly not a *Penicillium*. His preparations from subculture were some monoverticillate *Penicillium*. Conidia as reported were smooth and  $2\mu$  in diameter, but the cultures were lost.—Two preparations, one from the original material, one from a culture, remain in Lindner's collection and have been seen by us.

670. *P. repens* Cooke and Ellis. Grevillea 7, p. 6, 1878, type no. 553 in North American Fungi, Newfield, N. J., October, 1879. (See fig. 99.)

Colonies upon rotten wood floccose, forming brown cushions or masses, as seen in exsiccati; hyphae branching, septate, brown walled, about  $4\mu$  in diameter with cells 20 to  $30\mu$  long; conidiophore 7 to  $25\mu$  occasionally up to  $100\mu$  long, 1 to 2 or occasionally several septate, arising as branches perpendicular to aerial hyphae; conidial apparatus branching variously

3 to 4 times verticillate with primary series of branches when present 10 by  $4\mu$ , secondary series 5 to 7 by 3 to  $4\mu$ , metulae 5 to 7 by 3 to  $4\mu$  and sterigmata about 7 by 2 to  $3\mu$ , all elements having cell walls brown when mature; conidia globose 2 to  $3\mu$ , colorless, developed in gelatinous masses forming aggregates; suggesting *Gliocladium*.

The collection of the New York Botanical contains five packets of No. 553 in N. A. F., cited as type, collected at Newfield, New Jersey, by J. B. Ellis upon rotten wood and in addition, 1 packet upon magnolia, January, 1878, 3 packets upon magnolia in 1880, 1 packet upon rotten wood, 1885.

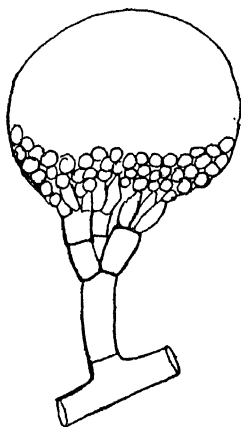


FIG. 99. *P. repens* of Cooke and Ellis: A penicillus from the exsiccati showed as a solid slime ball with determinable elements at the base upon which the diagnosis as *Penicillium* was based.

671. *P. saponis* B. and Br. No. 1913 of British fungi. Ann. Nat. Hist., 5th series, 7: 130, Pl. III, fig. 3. 1881.

Nigrum, monilibus e cellulis 2-3 oriundis; sporis globosis. On soap.

Synonym: *Haplographium saponis* (B. and Br.) Sacc.

672. *P. simplex* Lindner. Mikrosk. Betriebskontr., 1901, p. 315; Atlas Mikr. Grundl. Taf. 36; Saccardo Syll. XVIII, p. 518.

Synonym: *Scopulariopsis simplex* (Lindner) Vuillemin. Bul. Soc.

Mycol. France 27: 137-152, generic change, p. 143. 1911.

The species described as *P. simplex* in the 1901 edition, was later described as *Catenularia fuliginea* Saito and is so cited in the 5th edition

(1909), p. 385, of the same work. This species is cosmopolitan and well known to us in culture. Vuillemin (Bul. Soc. Mycol. France 27: 143, 1911) incorrectly placed this species in *Scopulariopsis* apparently upon purely bibliographic grounds.

673. *Penicillium sitophilum* Montagne. In Ann. Sci. Nat., ser. II, vol. XX, p. 377, pl. 16, fig. 4. 1843. This is the well-known *Monilia sitophila* (Mont.) Sacc., as indicated in Sylloge 4: 35. 1886.

674. *P. sparsum* Grev. Scottish Crypt. Flora, vol. 1, pl. 58, fig. 2a and b, 182.

Vegetative mycelium white, with scattered stalks bearing pure white heads of "sporules" visible to the naked eye.

Habitat: on semiputrid stems of herbaceous plants, where it forms whitish spots  $\frac{1}{2}$  to 1 inch long and several lines wide.

Culture: None.

A white form but probably not a *Penicillium* as indicated by Link, Sp. Plant., Ed. 4, vol. 6, pt. 1, p. 20, 1924. Berkeley, in British Flora (Hooker) V, pt. II, p. 344, 1836, cites *Monilia penicillata* Fries and *Briarea elegans* Corda as synonyms.

675. *P. verticillatum* Corda. Icones I, p. 21, table 6, fig. 281. 1837.

Stalk erect, simple, septate, branching at apex, branches with branchlets forming a head in alternately sub-verticillate fashion. Conidia in chains, simple or branched, ovate, white, 0.00017. Found on leaves of rotten begonia, near Prague, 1835.

Not a *Penicillium* in the sense of this paper, although Biourge gives it in his "List onomastique" as *P. (Spic.) verticillatum* 1837, Harz 1871. Harz appears to have used *Spicaria verticillata* (Corda) Harz.

675a. *P. violaceum* Biourge. In Semal. O. 1897.

The name was given in Biourge's list as attributed to himself but he failed to discuss it in his monograph and we have not found the description.

676. *P. viride* Fresenius. Beitr. z. Myk., p. 21-22, Taf. III, figs. 16-19.

Described from little gray green colonies on rotten grapes. From the figures and description given, this was some member of the *Cladosporium-Hormodendrum* series with conidiophores erect, branched, each

branch ending in dichotomous spore chains; spores elliptical or oval, mostly two guttulate. Relation to *Cladosporium* was recognized by Fresenius.

677. *P. viride* Rivolta. Parass. Veget., p. 453, Tav. VI, fig. 154a. 1873.

Rivolta recognized that this belonged with other species which were even then being placed in *Cladosporium* or *Hormodendrum* by others.

678. *P. viridum* Sopp. Monogr., pp. 198–200, Taf. XIX, fig. 137; Taf. XXIII, fig. 41. 1912.

Colonies upon meat-peptone-sugar-gelatine at first blue green, then becoming yellow-green; hyphae fairly fine or delicate; reverse white to yellow or chrome-yellow; odor pleasant, rooty; conidiophores coarse, rather short, irregularly branched and septate (jointed = "gegliedert"), figured with its cells each enlarging upward clavate, and as producing 1 or more divergent, clavate metulae at the first node back from the apex and a terminal verticil of 4 to 8 metulae at the apex, each metula producing a *Citromyces*-like group of sterigmata and parallel conidial chains, or some metulae in the same verticil occasionally branched to produce secondary metulae; conidia oval, small, increasing in size toward the end of the chain, when fully ripe, 7 by 5 to 6 $\mu$ .

Species found in soil. Cultures grew best at 20°, with minimum at 5° and maximum at 35°C., and grew well in milk, urine, broth, wort, potato, rice. Milk was coagulated with the separation of a greenish serum, and the production of reddish mycelium; rice was colored yellowish green. Conidia remained viable more than three years.

Careful reading of Sopp's description leads to the conclusion that this species was an *Aspergillus* and in all probability belonged in the group now typified by *A. conicus* Blochwitz. Two things are to be noted—the enlargement upward of the cells of the conidiophore and its branches, the conidia up to 7 by 6 $\mu$  enlarging toward the end of the chain.

## CHAPTER XXVII

### SPECIES, IDENTIFICATION, PROCEDURE, GROUP KEYS AND SPECIAL KEYS SPECIES

What then is a species? There is no answer which will satisfy all demands. From the standpoint of the culture laboratory, two organisms belong to the same species if they have the same morphology including both colony characters and microscopic characters, and agree in such routine reactions as are determinable without the introduction of elaborate quantitative analytical methods. The extent of variation to be tolerated in such a requirement if rigidly determined would call for repeated parallel culture of both organisms upon a wide range of substrata in succession and under successive variations of such factors as aeration, temperature, humidity, concentration and lighting, with comparative study of the variations induced by such changes of environment. The range of variability might then be tabulated for each organism and their identity or separateness established by comparison of the tables. Such a program is time consuming and clogs the records with masses of detailed data which are difficult to analyze. Yet such information fills many pages in our publications and still more remains unpublished in the files of our culture laboratories.

The specification above excludes the results of quantitative analysis as requiring more than the culture laboratory can comply with when large numbers of organisms are presented for study. We are faced then with such an incident as this: Thom in 1915 described *P. purpurogenum* var. *rubri-sclerotium* (no. 2670). In 1924, no. 4756 was isolated from moldy paper and exactly repeated the morphology and routine reactions originally found in no. 2670. Meantime Herrick and May had selected our stock culture descended from the original no. 2670 as the basis of a successful experimental production of gluconic acid. A culture of no. 4756 was given to them for comparison. It proved entirely worthless when subjected to the same tests as no. 2670 in their experimental work. Morphological identity with superficial similarity in routine reactions such as color production is not necessarily correlated with possession of other biochemical attributes in quantitatively equal measure. Identity biochemically as well as morphologically would require a quantitative study of the activities of each strain. That is not all, for enough obser-

variations exist to raise a doubt whether there is a biochemical *status quo* which can be established for every species in culture today and can be expected from successive transfers of the same strain over a period of five years. The gluconic-acid organism already discussed has certainly lost its original and conspicuous power of producing abundant green conidia, red sclerotia and intensely red colors in the substrata within a growing period of a few days. Other organisms are certainly more stable but such stability must be established strain by strain before it can be depended upon. The existence of great series of related strains such as those typified by *P. expansum*, *P. chrysogenum*, *P. roqueforti*, *P. terrestre* and others is evidence that mutability is the rule rather than the exception in the whole group.

The differences between these related strains are commonly described in terms of shades of colony color, of color in the substratum, mass of growth, and details of branching, measurements, and markings. The causes of these variations are unknown but enough has been done to suggest that a particular series of enzymes may be produced by all members of a series of organisms but in very different quantity strain by strain. If then a score of enzymes are represented in the complex, the possible number of permutations becomes almost endless.

The particular aspect of a species in laboratory culture at any particular time is then the response of that organism to the environment presented. But since that mold is growing upon a given volume of a substratum containing a mixture of several fermentable or decomposable materials, all other conditions may be held constant yet the nature of the nutrient substratum will change rapidly from day to day. For example, a substratum containing a fermentable sugar initially presented at pH 7, may quickly become as acid as pH 3, then by the gradual utilization of the acid or by the production of alkaline by-products from the decomposition of nitrogenous material also present in the substratum or by both combined, may again reach pH 7 or even pH 8 in a few weeks. The figures suggested are extreme but the change from neutral to acid and back to neutral or even alkaline conditions in the culture medium upon which a *Penicillium* is growing is usual, not exceptional. Under extreme conditions, changes in structure, form and measurement may also be extreme.

From a practical taxonomic standpoint, all obtainable information is valuable. Conspicuous reactions or structures regularly obtained under definable conditions are most useful. But to keep within the physical limits of the culture room and the time and energy of the worker, the broad lines of separation among many strains in *Penicillium* can be

established more readily by rigid comparative culture upon one or at most two or three culture media than by an attempt to define the all possible limits of variation strain by strain. Within the homogeneous group or series, detailed study upon elaborate series of media may be necessary to make identification of the individual strain even probable. For the purposes of this book, species are aggregates of strains with essentially the same morphology and agreeing in their qualitative reactions in culture media, with minor quantitative differences in reaction mostly ignored. The lines of such separation are based upon standardization of culture methods and media, upon standardization of the descriptive information sought and of the terms used.

To identify a species, therefore, three requisites must be furnished: (1) the specimen, (2) proper equipment, and (3) a standardized description.

#### IDENTIFICATION

Identification of a *Penicillium* requires consideration of (1) the specimen, (2) equipment needed (3) preparation of an orderly description, and may be facilitated by the use of (4) a group key or (5) special keys.

The taxonomic section (in this book, the "chapter") represented by a species can probably be reached by the occasional worker most easily by using the arbitrary group key, or, exceptionally by a key based on substrata or special characters; if material is to be studied frequently the analysis of groups in Chapter XI gives a better picture of the actual relationships. Within each group the choice between a section analysis and an arbitrary key is presented.

Cross-references between group keys are introduced in many cases where they are believed to be needed.

##### *1. Specimens, material for identification*

Identification of species of *Penicillium* must remain the task of the culture laboratory, rather than the microscopical worker in the herbarium. Material or collections received for identification may come in the form of pure cultures upon natural or artificial substrata in tubes, dishes, flasks or other containers. Such material is in favorable condition for transfer to fresh substrata; only occasionally is the culture received in condition for immediate study and identification. The widely known practice of sending such cultures by mail after purification by the finder, consists in lifting the colonies from a petri dish culture with a spatula, placing them upon sterile paper, folding the paper into a packet which



is dried in air until it can be slipped into an envelope for mailing. This has given generally satisfactory results with *Penicillia* over many years.

Moldy substrata such as pieces of fruit, vegetables, nuts, bark, bits of vegetation, enclosed in clean packets or even in envelopes and mailed, require the additional work of making the initial examination of raw material (with a hand lens preferably), making the primary cultures and the selection and purification of the species found before attempts at identification can be safely made. The difficulties in examining material sent in are well illustrated by a variant culture recently received as pure with its dominant *Penicillium* readily recognized except for an unexpected discoloration of the substratum. This was satisfactorily explained when on continued watching as our cultures became old, *Aspergillus sydowi* appeared at several places in the colony.

In our work with *Penicillia* we have used Czapek's solution agar (see Chapter V) for general study of species; it is readily prepared; it should not differ materially in different laboratories; its reaction is around pH 6.6 to 7; as prepared, it is colorless, hence furnishes a favorable opportunity to discover color changes due to mold growth; most species grow upon it, but only moderately well.

## 2. Equipment

### *Necessary:*

1. A good microscope with fairly high magnifications including the equivalent of a good oil immersion objective, and an accurately calibrated micrometer.
2. Pure cultures of the organism grown upon standard substrata.
3. Observations recorded over a growing period of about two weeks under favorable (recorded) range of temperature. These must include direct observation of the undisturbed growing colony with lower magnifications of the compound microscope.

### *Desirable:*

4. Color standards such as Ridgway's Nomenclature of Colors, or Klincksieck and Valette's Code de Couleurs.
5. Drawings of characteristic structures and sketches of habit.
6. An orderly description of the organism for which the following outline is suggested.

## 3. Outline of description indicating data essential

1. Substratum.
2. Margin broad, narrow, submerged, aerial, cobwebby.

3. Submerged hyphae coarse or delicate, color, approximate range of diameter in microns.
4. Colony color and color changes.
5. Reverse color and color changes.
6. Spreading or restricted in growth, zonation present, absent or slowly evident.
7. Floccose, velutinous, fasciculate, or funiculose, wrinkled, buckled, smooth or plane.
8. Odor.
9. Drops (transpired fluid).
10. Sclerotia, perithecia (details if any).
11. Conidiophores length, diameter, with walls smooth, rough, pitted, crusted, warty, arising from aerial or submerged hyphae (which ?) course or aggregation, branching.
12. Penicillus: length of branching system free from conidial chains, characteristic shapes, branches, number of series, arrangement, measurements.
13. Metulae, measurements, arrangement.
14. Sterigmata, measurements, arrangement, shape.
15. Conidial chains arranged—in columns, parallel, divergent or tangled.
16. Conidia—size, shape, color, markings.
17. Source and significance in nature and relation to special substrata. (See Keys).

#### 4. Artificial key to group names used as chapter headings

- A. Conidia thin walled, without a ring or collar at base...B.
- A. Conidia rather thick-walled, with a thickened ring or collar at the truncate base.....*Scopulariopsis*. Chapter XXIV.
- B. Conidia not in chains, and not enveloped in slime as seen in laboratory culture.....C.
- B. Conidial chains soon broken up and the conidia forming a mass or ball enveloped in slime as observed in cultures.....*Gliocladium*. Chapter XXII.
- C. Sterigmata in characteristic verticils with conidium producing points or tubes straight; conidial areas commonly some shade of green, blue green or gray green during the growing period; white, yellow or reddish forms occasional but few.....D.

- C. Sterigmata irregularly produced, partly variously arranged on fertile branches, partly in verticils, mostly tapering to long slender points which are commonly curved or bent from the main axis of the sterigmata, conidial areas never green (see Fig. 92)... *Paecilomyces*. Chapter XXIV.
- D. Penicillus or conidial apparatus characteristically monoverticillate. The individuality of the apical cell with its verticil of sterigmata each bearing a conidial chain and the whole constituting a monoverticillate penicillus is fairly sharply maintained whether the fertile hypha is unbranched or produces branches in more or less regular manner, each bearing at its apex a similar penicillus..... Monoverticillata Chapter XII and XIII.
- D. Penicillus more complex than monoverticillate..... E.
- E. Penicillus biverticillate consisting of two series of elements—sterigmata and metulae (or branchlets)..... G.
- E. Penicillus usually containing three or more series of elements, sterigmata, metulae and branches or branchlets, or if biverticillate mostly asymmetrical... F.
- F. Penicillia symmetrically polyverticillate consisting of at least three symmetrical and superposed series of elements..... Polyverticillata symmetrica. Chapter XXI.
- F. Penicilli with branching system characteristically forming in incomplete or one-sided verticils, whether in two series of in three or more series..... H.
- G. Penicillus biverticillate but with second verticil (metulae or branchlets) incomplete about the base of the main axis extended..... I.
- G. Penicillus biverticillate and symmetrical..... Biverticillata-symmetrica. Chapter XX.
- H. Penicilli when terminal usually biverticillate..... I.
- H. Penicilli mostly consisting of three or more series of elements..... K.
- I. Metulae divergent..... J.
- I. Metulae appressed (usually one or more elements of the third verticil present)..... *Brevi-compacta*. Chapter XV.
- J. Colonies floccose..... *Lanata-divaricata*. Chapter XVII.
- J. Colonies funiculose..... *Asymmetrica-Funiculosa*. Chapter XVIII.

- J. Colonies velvety.....*Velutina-divaricata*.  
Chapter XIV.
- K. Conidiophores borne separately (not in fascicles or coremia).....L.
- K. Conidiophores partly or entirely produced in fascicles or coremia.....Fasciculata. Chap-  
ter XIX.
- L. *Penicillus* short, with elements compact at base with tips and conidial chains divergent. Colonies ranging from lanose to velutinous.....*Brevi-compacta*.  
Chapter XV.
- L. Colonies lanose or floccose.....*Lanata-typica*.  
Chapter XVI.
- L. Colonies velutinous.....*Asymmetrica-Velutina*. Chapter  
XIV.

### 5. Certain conspicuous characters

Certain conspicuous characters or occurrence upon particular substrata form a presumptive basis for quick identification of single species or small groups of species often widely scattered through the whole penicillate series. Some of these characters apparently appear independently in the several sections of the *Penicillia*, hence may be usefully indicated here as well as reached in subsequent analyses of those sections. The determination can usually be made with low magnifications of the compound microscope.

The following observations have been selected as practically useful and lead to numbered paragraphs in this chapter.

	<i>Paragraph</i>
Production of sclerotia.....	1
Production of perithecia and ascospores.....	2
Production of coremia.....	3
Production of superficial ropes of hyphae.....	4
Colonies white (never green).....	5
Colonies salmon to pinkish.....	6
Colonies ochraceous to avellaneous.....	7
Colonies in lilac, violaceous or hyacinth colors.....	8
Species associated with particular substrata. Cheese.....	9
Apples, pears, grapes.....	10
Citrus fruits.....	11
1. Sclerotium Producing Species.	
Sclerotia associated with <i>Monoverticillata</i> .....	Chapter XII. 110
Sclerotia associated with <i>Biverticillata-symmetrica</i> .....	Chapter XX. <i>P. ki- liense</i> .

Sclerotia associated with Asymmetrica.....	Chapter XIX. <i>P. gladioli. P. italicum. P. glaucum.</i>
Sclerotia associated with Scopulariopsis conidial forms.....	Chapter XXIV.
2. Perithecia and Ascospores Produced.	
Ascosporic forms associated with species of Biverticillata-symmetrica .....	Chapter XX.
Ascosporic forms associated with species of Asymmetrica .....	Chapter XIX. <i>Fasciculata.</i>
a. Conidia elliptical, glaucous.....	<i>P. italicum</i> , 328.
b. Conidia globose, green or glaucous .....	<i>P. glaucum</i> of Brefeld 577a.
c. Conidia white.....	<i>P. candidum</i> of Morini 557a.
Ascosporic forms associated with conidial forms of Scopulariopsis.....	Chapter XXIV.

The ascosporic and sclerotium producing *Penicillia* so far as described are limited to small sections in certain large groups to which they may be traced by this key.

3. Coremium producing types.
  - A. Species fruiting typically by coremia (erect and definite).
    - a. Coremia long (3-15 mm.).
      1. Conidial masses strictly terminal, olive green, fragrant.....*P. claviforme*. XIX.
      2. Upper third of coremia fertile, conidia green.....*P. duclauxi*. XX.
    - aa. Coremia small.
      1. Coremia definite, densely crowded, colony orange below.....*P. granulatum*. XIX.
      2. Coremium character indicated in cultures by clustering of conidiophores, definite coremia only in old cultures, becoming large and definite upon apples.....*P. expansum*. XIX.
4. Ropes of aerial hyphae (funiculose species):
 

Aerial hyphae partly aggregated into trailing or ascending (not or rarely erect) commonly branching and interlacing ropes or fascicles (funiculose).

  - a. Ropes or fascicles associated with the Scopulariopsis type of conidium formation.....Chapter XXIV.
  - b. Ropes or fascicles associated with conidial masses enveloped in slime.....*Gliocladium*. Chapter XXII.

- c. Ropes or fascicles associated with Monoverticillate penicilli.....Monoverticillata-stricta. Chapter XII.
- d. Ropes or fascicles associated with Asymmetriconfunisulosa.....Chapter XVIII.
- e. Ropes or fascicles associated with biverticillate penicilli.....Biverticillata. Chapter XX.
- f. Ropes or fascicles associated with polyverticillate penicilli.....Polyverticillata-symmetrica. Chapter XXI.
5. Colonies white.
- Conidia of the Scopulariopsis type—no. 2.....XXIV. *P. brevicaulis* var. *album* and *glabrum*. *P. costantini*. Bainier.
- Conidia and sterigmata of the Paecilomyces type.....Paecilomyces. XXIII.
- Conidial masses enveloped in slime.....Gliocladium. XXII.
- Conidial mass of the true penicillium type.
1. Conidia large elliptical.....Chapter XIV.
- Conidia 9 by 18 to 20 $\mu$ .....*P. niveum* Sopp.
- Conidia 4 to 7 by 6 to 8 $\mu$ .....*P. digitatum* var. *californicum*.
2. Conidia globose, colonies floccose.....*P. caseicolum* Bainier. Chap. XVI.
3. Conidia subglobose 3 $\mu$ , irregularly biverticillate.....*P. Braziliense* 420.
6. Colonies salmon to pinkish salmon.
- Conidia on mucilaginous masses.....Gliocladium (and possibly Clonostachys). Chapt. XXII.
- Conidia in chains as in the true Penicillia.....*P. vermoensi* Bionourge. XXII.
7. Colonies ochraceous to avellaneous.
- Avellaneous to brown with Scopulariopsis type of conidia.....Chapter XXIV.
- Yellowish ochraceous to avellaneous, with the Paecilomyces type of sterigmata and conidia.....Paecilomyces. Chapter XXIII.
- Avellaneous with biverticillium type of conidial apparatus.....*P. avellaneum* and *P. gilvum*. Chapter XX.

- Olive to ochraceous even to reddish brown  
 Chapter XVI.....*P. ochraceum* and  
 allies. Bainier.  
 Chapter XVI.
8. Colonies in lilac, violaceous, or hyacinth shades (not  
 green at any stage of growth).....*P. lilacinum* series  
 Chapter XVII.
9. Cheese molds.  
 Cheese (Camembert and Brie):
1. Floccose, white unchangeable, no odor.....*P. caseicolum* Bainier.
  2. Floccose, white to gray-green, no odor.....*P. camemberti*.
  3. Powdery, yellowish white, spores smooth, ammoniacal odor.....*P. brevicaulis* var.  
*glabrum*.
  4. Powdery, yellowish white, spores tuberculate, ammoniacal odor.....*P. brevicaulis* var.  
*album*.
  5. Forming yellowish-brown areas, spores rough, ammoniacal odor.....*P. brevicaulis*.
- Cheese (Roquefort):
1. Green streaks inside the cheese.....*P. roqueforti*.
10. Penicillium rot of apples, pears, grapes, etc.  
 1. Blue-green colonies finally producing coremia.....*P. expansum*.
11. Molds of citrus fruits.  
 1. Colonies of mold, blue-green.....*P. italicum*.
2. Colonies of mold, olive-green.....*P. digitatum*.

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*P. cyaneo-fulvum* Biourge, 267, 239; 259.  
*P. cyaneum* (Bainier and Sartory), 226, 164, 226; 182, 57.  
*P. cyclopium* Westling, 384, 375; 86, 113, 145, 419, 424.  
*P. daleae* Zal., 360, 358.  
*P. decumbens* Thom, 197, 167, 195, 229.  
*P. (Spicaria) decumbens* (Oudem. and Kon.) in Biourge's List.  
*P. deformans* Sopp, 558, 162; 340.  
*P. delacroixii* Sacc. in Biourge's List.  
*P. (Haplogr.) delicatum* (B. & Br.) in Biourge's List.  
*G. deliquescens* Sopp, 507, 506, 502, 510.  
*P. (Gliocl.) deliquescens* (Sopp) in Biourge's List.  
*P. (Spicaria) densa* (Giard) (Vuill.) in Biourge's List.

- Corollium dermatophagum* Sopp, 548, 545.
- P. (Coroll.) dermatophagum* (Sopp) in Biourge's List.
- P. desciscens* Oud., 491, 444; 993.
- P. (Isaria) destructor* (Delacroix), 434, also in Biourge's List.
- P. dierckxii* Biourge, 206, 169; 393.
- P. (Stys.) difformis* (Oudemans) in Biourge's List.
- P. digitatoides* Peyronel, 245.
- P. digitatum* Sacc., 242, 236, 600; 18, 19, 76, 85, 125, 144, 246, 418, 532, 564.
- P. digitatum* var. *californicum*, 245, 235, 236, 599; 76.
- P. digitatum* var. *discoideum* March, 246, 236.
- Spicaria divaricata* Abbott, 544.
- P. divaricatum* Thom, 544, 15, 20, 29, 68, 73, 106, 110, 120, 129, 131, 133, 137, 134.
- P. divaricatum* Thom-Biourge, 530.
- P. divergens* B. & S., 427, 379; 57, 381, 414.
- P. "dubiosum* Wehmer," 559.
- P. duclauxii* Delacr., 458, 440, 598; 49, 56, 60, 82, 101, 102, 107, 417, 438, 145.
- P. duponti* G. & M., 248, 236.
- P. echinatum* Rivolta, 580.
- P. (Hapl.) echinatum* (Rivolta) (Sacc.) in Biourge's List.
- P. echinatum* Dale, 351, 162, 168; 338, 340, 350, 438.
- P. echinulatum* Dale, 353.
- P. (Corem.) elasticae* (Koorders) in Biourge's List.
- P. elegans* Sopp, 470, 441, 467.
- P. elegans* Corda, 549.
- P. (Isaria) elegantula* in Biourge's List.
- P. elongatum* Bainier, 485, 20.
- P. elongatum* Dierckx, 411, 378; 405, 409.
- P. epigeum* B. & C., 559.
- P. epigaeum* B. & C., 559, 514.
- P. epsteinii* Lindau, 312, 307; 310.
- P. erectum* Bainier, 295, 291.
- Paecilomyces erectus* Demelius, 486, 444, 546.
- P. (Isaria) eriopoda* (Bainier) in Biourge's List.
- P. exiguum* Bainier, 492, 444, 236.
- C. exiguus* Bainier and Sartory, 580.
- P. expansum* Link, 402, 377, 380, 598, 600; 1, 25, 49, 72, 76, 85, 89, 101, 102, 111, 123, 126, 132, 317, 388, 402, 423, 459, 561.
- P. expansum* Thom, 406.
- P. (Isaria, Spic.) farinosa* (Vuill.) in Biourge's List.
- P. fasciculatum* Sommerfelt, 559.
- P. fastigiatum*, 465, 441.
- P. favedkamp* "Kap" culture listed by Pribram, 213, 170.
- P. (Isaria, Spic.) felina* (D. C.) (Vuillemin) in Biourge's List.
- P. fellutanum* Biourge, 198, 167, 237; 436.
- P. fieberi* Corda, 560, 68, 219.
- Glio. fimbriatum* G. & A., 508, 509.
- Spicaria fimetaria* Moesz, 549, 545.
- P. (Stys.) fimetarius* (Karsten) in Biourge's List.
- S. fimicola* (Cost. & Matr.) Vuillemin, 531.
- P. (Corem.) fimicola* (Marchal) in Biourge's List.
- P. finitimum* Preuss, 580.
- P. (Haplogr.) finitimum* (Preuss) in Biourge's List.
- P. firmum* Preuss, 580.
- P. flavido-marginatum* Biourge, 314, 307.
- P. flavi-dorsum* Biourge, 224, 171, 172.
- P. flavo-cinereum* Biourge, 184, 165.
- P. flavo-fuscum* Biourge, 560.
- P. flavo-glaucum* Biourge, 386, 375, 241.
- P. flavo-virens* Cooke and Massee, 581, 443.
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- P. (Acaul.) flavum* (Sopp) in Biourge's List.

- P. flavum* Marchal, 550, 235, 545.  
*P. flexuosum* Dale, 419, 378, 418, 436.  
*P. flexuosum* Preuss, 581, 580, 419.  
*P. flexuosum* Westling in Biourge's List.  
*P. fluitans* Tiegs, 185, 165.  
*C. fôtens* Sopp, 213, 171.  
*P. frequentans* Westling, 216, 102, 131, 133, 137, 217, 218, 221, 387.  
*P. fuliginea* (Saito) in Biourge's List.  
*Acaulium fulvum* Sopp, 531, 514.  
*P. (Rhodoceph.) fulvum* (Rabenh.) in Biourge's List.  
*P. fulvum* Rabenhorst, 581.  
*Acaulium fulvum* Sopp, 531.  
*P. (Aspergillopsis) fumosus* (Sopp) in Biourge's List.  
*P. funiculosum* Thom, 464, 441; 131, 132, 466.  
*P. fuscipes* Preuss, 581, 580.  
*P. fusco-glaucum* Biourge, 325, 309.  
*C. fuscus* Sopp, 180, 163.  
*P. geophilum* Oud., 220, 172.  
*P. gilmanii* Thom, 345, 330.  
*P. gilvum* Sopp, 454, 443, 439, 599.  
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*C. glaber* Wehmer, 187, 105, 220.  
*P. glabrum* series, 215, 171.  
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*P. gladioli* Machacek, 381, 375, 598; 85, 113, 416.  
*P. (Corem.) glandicola* (Oudemans) in Biourge's List.  
*Floccaria glauca* Greville, 402, 409, 377.  
*P. glauco-ferugineum* Sopp, 364, 359; 145.  
*P. glauco-griseum* Sopp, 346, 237, 330, 340.  
*P. glauco-ochraceum* Preuss, 560.  
*P. glauco-ochraceum* Sopp, 560 in Biourge's List.  
*P. glauco-roseum* Demelius, 344, 330.  
*Coremium glaucum* Link, 25, 124, 404, 431.  
*C. glaucum* var. *fimicola* Marchal, 433.  
*P. glaucum* Link, 2, 560, 598; 1, 10, 26, 72, 74, 77, 82, 84, 87, 93, 95, 111, 114, 126, 132, 139, 147, 152, 264, 277, 374, 397, 402.  
*P. glaucum* Link, Wehmer, 407, 377.  
*P. glaucum* var. *coremium* Sacc., 406.  
*P. glaucum* var. *crustaceum* Fr., 562.  
*P. glaucum* var. *epimyces* Sacc., 561.  
*P. glaucum* var. *epixylon* de Thümen, 563.  
*P. glaucum* var. *fasciculatum* Pers. or B, 559.  
*P. glaucum* var. *fôtidum* Sopp, 56.  
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*P. gliocladioides* Preuss, 563, 163.  
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*P. (Stysanus) globosa* (Peglion) in Biourge's List.  
*P. godlewskii* Zal., 365, 359.  
*P. gonorrhoeicum* Hallier, 581, 572, 130.  
*P. (Trichurus) gorgonifer* (Fairier) 382 and in Biourge's List.  
*P. gorgonzola* Weidemann, 284, 241.  
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*P. granulatum* Bainier, 429, 379, 598; 61, 101, 414, 417, 424, 427.  
*P. gratioli* Sartory, 338, 330; 85, 99.  
*P. griseo-atrum* Biourge, 582, 169.  
*P. griseo-bruneum* Sopp, 287, 241.  
*P. griseo-brunneum* Dierckx, 302, 292.  
*P. griseo-fulvum* Dierckx, 371, 370, 360; 272, 557, 564.  
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*P. griseo-roseum* Dierckx, 263, 239; 109, 259.  
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*P. griseum* Bonorden, 563.  
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*P. guegueni* Biourge, 583.  
*P. guttulosum* Abbott, 343, 330; 201, 132.  
*P. hagemi* Zal., 298, 292.  
*P. herquei* B. & S., 467, 468, 441; 20, 57, 107, 469.  
*P. (Vertic.) heterocladum* (Penzig) in Biourge's List.  
*P. hickeyi* Biourge, 583.

- P. (Corem.) hiemale* (Bonorden) in Biourge's List.  
*P. hirsutum* B. & S., 493, 444, 440, 564.  
*P. hirsutum* Dierckx, 425, 379; 113, 356, 425.  
*P. howardii* Thom, 368, 359.  
*P. humicola* Oud., 444, 441; 66, 463, 564.  
*P. (Gliocoph.) hyalina* (Matruchot) in Biourge's List.  
*P. hypomyces* Biourge, 583, 191.  
*P. hypomyces* Sacc., 584.  
*P. implicatum* Biourge, 210, 170.  
*P. implicatum* Biourge var. *aureo-marginatum* Thom, 211, 170.  
*P. incarnatum* Berk., 565.  
*P. insectivora* (Sopp) Biourge, 532.  
*P. insectivorum* Sopp, 532, 514.  
*P. (Acaul.) insectivorum* (Sopp) in Biourge's List.  
*P. insigne* Bainier, 495, 557.  
*P. insigne* Sacc., 519.  
*P. insigne* (Winter) Schröter, 504, 34, 519.  
*P. intricatum* Thom, 367, 359; 365, 366.  
*P. islandicum* Sopp, 466, 441; 204.  
*P. italicum* Wehmer, 412, 378, 375, 598, 600; 61, 68, 74, 85, 86, 119, 125, 126, 152, 244, 246, 374, 414.  
*Scop. ivorensis* Boucher, 533, 142.  
*P. janczewskii* Zal., 354, 331; 249, 350, 360.  
*P. janthinum* Biourge, 341, 330, 359; 338, 361, 365.  
*P. jantho-citrinum* Biourge, 208, 169.  
*P. janthogenum* Biourge, 387, 376; 308.  
*P. jenseni* Zal., 346, 330, 226.  
*P. johannioli* Zal., 391, 376, 389.  
*P. juglandis* Weidemann, 416, 378; 128, 409.  
*P. "Kap laboratorium* Sopp," 407.  
*P. kapuscinskii* Zal., 355, 331.  
*P. kiliense* Weidemann, 455, 439, 597.  
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*P. krzemieniewskii* Zal., 362, 358, 360.  
*P. lacteum* Centralbureau List, 1929.  
*C. lacticus* Mazé and Perrier, 190, 166.  
*P. lacticus* Oertler in Biourge's List?  
*P. lagerheimii* Westling, 490, 444; 316.  
*P. lanoso-coeruleum* Thom, 322, 308.  
*P. lanoso-grisellum* Biourge, 246, 236.  
*P. lanoso-griseum* Thom, 327, 309.  
*P. lanoso-viride* Thom, n. sp. 314, 307.  
*P. lanosum* Westling, 317, 308.  
*P. lemoni* Sopp, 469, 441; 340.  
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*P. leucocephalum* Rabh., 585.  
*P. leucopus* (Pers.) Biourge, 408, 378; 11, 402.  
*P. (Gliocl.) lignicolum* (Matruchot) in Biourge's List.  
*G. lignicolum* Grove, 510.  
*P. lilacinum* Thom, 331, 329, 600; 73, 94, 131.  
*P. linguae* Panayotatou, 533, 143.  
*P. lividum* Westling, 205, 168; 192.  
*P. lobulatum* Bonorden, 585.  
*P. (Gliocl.) luteolum* (Von Höhnel), 509, and in Biourge's List.  
*P. luteo-viride* Biourge, 461, 441; 200.  
*P. luteum* Zukal, 448, 442, 439; 14, 49, 56, 74, 82, 104, 108, 131, 132, 137, 154, 445.  
*P. luteum* series, 479, 555.  
*P. luteum* Sopp, 443.  
*P. maculans* Sharples, 565, 130.  
*G. macropodium* El. Marchal, 509.  
*P. macrosporium* B. & Br., 565.  
*P. majusculum* Westling, 389, 376, 387.  
*P. malivorum* Ciferri, 409, 378; 123.  
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*P. martensii* Biourge, 388, 376.  
*P. matris-meae* Zal., 349, 330, 444.  
*P. maydis* Lewin, 565.  
*P. (Stys.) media* (Sacc.) in Biourge's List.  
*P. (Gliocl.) megalosporum* (Marchal) in Biourge's List.  
*P. megalosporum* B. & Br., 566.  
*P. meleagrinum* Biourge, 266, 239.  
*P. melinii* Thom, n. sp., 273, 240.  
*P. microsporium* Riv., 566.  
*P. miczynskii* Zaleski, 488, 444.

- P. minimum* Siebenmann, 566, 138.  
*P. minio-luteum* Dierckx, 464, 441.  
*C. minutus* B. & S., 229, 164, 226; 200, 57.  
*P. (Isaria) monilioides* (Albert. & Schw.) in Biourge's List.  
*P. monstrosum* Sopp, 566.  
*P. montoyai* Castellani, 567, 141.  
*P. morsus-ranae* Corda, 567, 169.  
*P. multicolor* G.-M. & P., 212, 170.  
*C. musae* B. & S., 228, 163, 226; 57.  
*P. musae* Weidemann, 393, 376; 229.  
*P. mycetogenum* Negri, 567.  
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*P. mycetomi* Neveu-Lemaire, 568, 140.  
*P. namyrowskii* Zaleski, 484, 443.  
*P. (Corem.) necans* (Oudemans) in Biourge's List.  
*P. (Corem.) necans* (Fischer) in Biourge's List.  
*P. (Gliocl.) nicotianae* (Oudem.), 509 and in Biourge's List.  
*P. nigrescens* Junghuhn, 431, 379.  
*P. (Corem.) nigrescens* (Junghuhn) in Biourge's List.  
*P. nigricans* Bainier-Thom, 351, 331, 168; 350, 360.  
*P. nigrovirens* Fres., 585.  
*Acaulium nigrum* Sopp, 534, 514.  
*P. niklewskii* Zal., 193, 167.  
*P. niveo-rubrum* Sopp, 457, 440.  
*P. (Corem.) niveum* (Corda) in Biourge's List.  
*P. niveum* Bainier, 497.  
*P. niveum* Sopp, 242, 235, 599.  
*P. notatum* Westling, 264, 239; 131, 137, 259, 262, 391.  
*P. obscurum* Biourge, 251, 237; 255.  
*P. (Spicar.) ochracea* (Boudier) (Vuillemin.) in Biourge's List.  
*P. (Oospora) ochracea* (Libert) (Sydow) in Biourge's List.  
*P. ochraceum* Bainier-Thom, 309, 306, 600; 133.  
*P. ochraceum* var. *macrosporum* Thom, 310.  
*P. ochro-chloron* Biourge, 363, 359.  
*P. ochroleucum* Artault, 568.  
*P. oidioforme* Orlova, 585.  
*P. oledzkii* Zaleski, 221, 172.  
*C. olivaceus* Sopp, 179.  
*P. olivaceus* Sopp, 31, 242.  
*P. olivaceum* Corda, 586.  
*P. olivaceum* Wehmer, 245; 236; 125, 242, 550.  
*P. olivaceum* var. *discoideum* Marchal, 586, 246.  
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*P. (Isaria) ophioglossoides* (De Stephani) in Biourge's List.  
*P. (Oospora) opoixi* (Delacr.) in Biourge's List.  
*P. orbicula* Corda, 586.  
*P. (Briarea) orbicula* Corda in Biourge's List.  
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*P. ovoideum* Preuss, 586.  
*C. oxalicus* Maze and Perrier, 190.  
*P. oxalicum* Currie and Thom, 247, 236; 82, 115, 125, 398, 574.  
*P. paczoskii* Zal., 202, 168.  
*P. palitans* Westling, 396, 376; 100, 113, 145.  
*P. pallido-fulvum* Peck, 568.  
*P. parasiticum* Sopp, 455, 439, 454, 554.  
*P. patris-mei* Zal., 303, 292.  
*P. patulum* Bainier, 420, 379; 294, 517.  
*P. paxilli* Bainier, 294, 291; 21, 289, 295, 421.  
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*P. (Gliocl.) penicillioides* (Corda-Matruchot) in Biourge's List.  
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*P. (Monilia) penicillioides* (Delacroix, 535, 146, 514.  
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- P. petchii* Sartory and Bainier, 454, 453, 439; 154.  
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*P. phaeoanthinellum* Biourge, 200, 183.  
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*P. piscarium* Westling, 487, 444, 359.  
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*P. platense* Spegazzini, 196, 167.  
*P. plicatum* Bonorden, 569.  
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*P. psittacinum* Thom, 369, 360, 309; 315, 392.  
*P. (Spicaria) psychidae* (Evans) in Biourge's List.  
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*P. puberulum* Bainier, 271, 240; 103, 107, 269, 288, 318, 421.  
*P. (Cit.) purpurascens* Sopp (Misprint) in Biourge's List.  
*P. purpurogenum* Fleroff-Stoll, 478.  
*P. purpurogenum* Stoll, 478, 442; 93, 107, 436.  
*P. purpurogenum* var. *rubri-sclerotium* Thom, 479, 442; 51, 122.  
*C. purpurescens* Sopp, 178, 163.  
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*P. racemosum* Hoff-Westendorp, 587.  
*M. racemosum* Pers., 587.  
*P. raciborskii* Zal., 318, 308.  
*P. radians* Bonorden, 570.  
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*P. (Stysanus) ramifer* (Rolland) in Biourge's List.  
*P. ramosius* Grov., 572.  
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*P. repandum* Bainier and Sartory, 550, 545.  
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*P. repens* Cooke and Ellis, 588, 510.  
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*P. rivolii* Zal., 342, 330, 358.  
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*P. rogeri* Wehmer, 310.  
*P. rolfssii* Thom, 489.  
*P. roquefort* Sopp, 286, 241.  
*P. roqueforti* Thom, 277, 240, 600; 50, 56, 57, 72, 76, 79, 83, 87, 89, 94, 101, 102, 117, 119, 120, 125, 132, 273.  
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*P. rosato-fragens* Biourge, 570.  
*P. (Oospora) rosatum* Biourge, 537, 514.  
*P. roseo-cinnabarinum* Biourge, 204, 163, 168.  
*P. roseo-citreum* Biourge, 323, 308; 259, 391.  
*P. roseo-griseum* Biourge, 264.  
*P. roseo-maculatum* Biourge, 186.  
*P. roseo-purpureum* Dierckx, 181, 164.  
*P. roseum* Link, 504, 509, 18, 108, 121, 129, 266, 500.  
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*P. (Gliocl.) roseum* (Matruchot) in Biourge's List.  
*Gliocladium roseum* Bainier, 502, 504; 509.  
*P. (Vertic.) roseum* (Cooke) in Biourge's List.  
*P. roseum* (deKral) Thom in Biourge's List.  
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*P. rugulosum* Thom, 472, 442, 441; 108, 125, 127, 131, 354, 437, 471, 479.  
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*P. (Oospora) salina* (Nomyslowsky?) in Biourge's List.  
*Citromyces sanguifluus* Sopp, 180, 164.  
*P. sanguineum* Sopp, 476, 442; 478.  
*P. saponis* B. & Br., 588.  
*P. sartoryi* Thom, 233, 226.  
*P. schneggii* Boas, 417, 378, 236, 375; 49.  
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*P. (Spic.) silvatica* (Oudem. & Kon.) in Biourge's List.  
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